Studies on the clinical genetics of cancer

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R.H. Sijmons

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Stellingen behorende bij het proefschrift van R.H.Sijmons, Groningen, 7 juli 1999.

1) Erfelijke kanker bestaat niet

2) Het ondergaan van pre-symptomatisch DNA onderzoek dient geen voorwaarde te zijn voor het deelnemen aan periodieke controles

3) De familieanamnese voor kanker is niet zondermeer betrouwbaar
(dit proefschrift)

4) Artsen dienen bij patiënten met de ziekte van Hirschsprung alert te zijn op multipele endocriene neoplasie type 2A
(dit proefschrift)

5) Overgangsepitheelcarcinomen van nierbekken en ureter behoren tot het tumorspectrum van hereditair non-polyposis colorectaal carcinoom
(dit proefschrift)

6) Het nauwkeurig voorspellen van de klinische gevolgen van de aanleg voor een erfelijke tumorsyndroom is onmogelijk

7) Het is vanuit klinisch genetisch standpunt ongewenst dat de wet slechts de invuller van een overlijdenscertificaat later voor klinische doeleinden toegang tot dat document verleent

8) De preoccupatie binnen de genealogie met de paternale overerving is genetisch gezien misplaatst

9) Als een promotie een bevalling is, dan is het schrijven van de inleiding niet zelden de inleiding

10) Sommige ouders kunnen er niet van dromen dat hun kinderen 's nachts doorslapen

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`It's a Snark!' was the sound that first came to their ears,

And seemed almost too good to be true.

Then followed a torrent of laughter and cheers:

Then the ominous words, `It's a Boo---'

Then, silence. Some fancied they heard in the air

A weary and wandering sigh

That sounded like `--jum!' but the others declare

It was only a breeze that went by.

(*From: Lewis Carroll, The Hunting of the Snark, 1876*)

Voor Kirsten en mijn Vader

Contents

Aim of this thesis

Knowledge of clinical genetic aspects of cancer is rapidly accumulating. Still, many of these aspects are unclear. They range from differential diagnosis and DNA testing to preventive options and psychosocial issues. The studies reported in this thesis aim at contributing to the clarification of some aspects and focus on clinical recognition of human cancer syndromes, tumour risks and disease spectrum.

Chapter 1
Introduction

1.1 Definitions:

Clinical genetics is the medical discipline devoted to the diagnosis of hereditary disorders and the counselling of individuals and families confronted with these disorders.

The term *consultand* (as opposed to consultant) has been introduced to refer to individuals seeking genetic counselling and testing[1-3]. Alternative terms are *consultee* and *counsellee*.

The *clinical genetics of cancer* is the clinical genetics of familial and hereditary cancer. The terms *clinical cancer genetics* and *clinical oncogenetics* are also used to refer to this particular application, which should be differentiated from the clinical use of testing for non-inherited (*i.e. somatic*) genetic changes in cancer (*e.g.* the testing for the presence of a rearrangement of the bcr and abl oncogenes in chronic myeloid leukaemia).

Genetic counselling is the communication process which deals with human problems associated with the occurrence, or risk of an occurrence, of a genetic disorder in a family[4].

Inherited cancer predisposition refers to inherited mutations (*germline mutations)* of a range of genes associated with an increased risk to develop particular types of cancer. In an unknown, but possibly quite substantial, proportion of all cancer cases, such a predisposition plays a role in cancer development. However, in probably the majority of cases this predisposition has only has a small to moderate effect on cancer risk. The best known examples are the wide range of hereditary variants of enzymes involved in (pro)carcinogen metabolism[5-8].

The term *hereditary cancer* is used in two different meanings. First, it is used to refer to cancer, developed in the presence of a strong inherited cancer predisposition. The level of life-time risk to develop cancer is a measure for strength of this predisposition, but at what level of risk a predisposition is classified as 'strong' has not been defined.

Second, *hereditary cancer* is used to refer to cancer which has developed in the setting of the *human cancer syndromes* (see below). The association between a particular cancer type and a human cancer syndrome is defined by the significantly more frequent occurrence of that cancer in carriers of the mutant gene than in a control population (*i.e.* increased relative risk). An increase in relative risk does not necessarily imply high life-time risk. Therefore, the first and second meaning of *hereditary cancer* are not identical. For example, ovarian cancer risk is significantly increased in female carriers of mutant mismatch repair genes (associated with hereditary non-polyposis colorectal cancer, HNPCC). Since the life-time risk to develop ovarian cancer in HNPCC is approximately 10 %[9] (compared with 1.5 % in the general population), the vast majority of carriers will never develop that tumour. Therefore the predisposition for ovarian cancer in HNPCC cannot be regarded as 'strong'.

Although *familial cancer* is sometimes used as a synonym of hereditary cancer, it is more appropriately used to simply refer to the occurrence in a family of more than one case of cancer, not necessarily hereditary in nature.

Non-hereditary cancer is defined as cancer, developed in the absence of a strong inherited cancer predisposition. Again, no cut-off risk levels have been defined. The term *sporadic* cancer refers to single cases of cancer in a family. Although it is widely used to refer to *non-hereditary cancer*, some of these single cases may very well be associated with a strong inherited cancer predisposition (cf. chapter 2, section 2.2.1).

Human cancer syndromes, also referred to as hereditary cancer syndromes or *familial cancer syndromes,* are those hereditary disorders for which the occurrence of (particular types of) cancer is considered a clinical hallmark. Again, there is no strict criterion to classify a feature as a hallmark. The term *hereditary cancer predisposition syndromes* is sometimes used as a synonym of human cancer syndromes, although the name suggests that it refers to hereditary disorders associated with any level of increase of cancer risk. By definition, all patients with hereditary cancer have a human cancer syndrome.

The disorders commonly included under the heading of the human cancer syndromes are listed in chapter 2 and more extensively in the Familial Cancer Database accompanying chapter 5.

1.2 Clinical genetics of cancer.

1.2.1 The genetic diagnosis

Central to the genetic analysis of a given family, is the question whether or not the family meets clinical and/or molecular criteria of one or (sometimes) more of the human cancer syndromes.

The first step in this analysis is taking the family history and drawing a pedigree based on this history. Because the family history is not necessarily accurate, it needs to be verified by checking medical records in order to make a correct interpretation.*(The accuracy of family history is further explored in chapter 3.)* Based on the (verified) pedigree data, a preliminary and sometimes definite (differential) diagnosis is made. *Clinical genetic differential diagnosis and software to help with that process, are discussed in chapters 2 and 5, respectively.*

Depending on the differential diagnosis, a physical examination of affected family members to check for dysmorphisms (*e.g.* macrocephaly in Cowden syndrome) or other physical characteristics (*e.g.* retinal hamartomas in von Hippel-Lindau disease) of human cancer syndromes may be appropriate. The same may be true for biochemical analysis (*e.g.* persistent HbF in Fanconi anaemia), cytogenetic analysis (*e.g.* in the chromosome breakage disorders or when a microdeletion syndrome is suspected) and DNA analysis. The number of hereditary disorders for which DNA analysis is possible, is rapidly growing. As yet, DNA analysis for a particular disorder may fail to detect a germline mutation, because the disorder may be genetically heterogenous and the mutation may therefore be present in a gene other than the one(s) tested, or because the test for a gene does not detect all possible mutations in that gene. DNA analysis for germline mutations in the genes associated with the human cancer syndromes is preferably performed in DNA extracted from normal cells (white blood cells, fibroblasts or sometimes paraffin blocks/frozen tissue) from affected relatives. DNA analysis of tumour tissue may sometimes also be informative, the classical application being the testing for microsatellite instability, typical for hereditary non-polyposis colorectal cancer (HNPCC)(*an example is presented in appendix IV*).

1.2.2 Genetic counselling

The process of genetic counselling aims at helping the individual and/or family to comprehend the medical facts, to appreciate the way heredity contributes to the disorder, and the risk of recurrence in specified relatives, to understand the alternatives for dealing with the risk of (re)occurrence (including reproductive options), to choose among alternative courses of action, and to make the best possible adjustment to the diagnosis of the disorder in an affected family member and/or to the risk of recurrence of that disorder[10]. Most of these issues are discussed after the genetic diagnosis has been made (genetic counselling part 2 in figure 1). However, psychological distress related to the family history, expectations on the side of the consultand as well as on the side of the geneticist with respect to the outcome of the genetic analysis and possible implications for the consultand's relatives are addressed during the intake (genetic counselling part 1 in figure 1).

Medical facts about the disorder: phenotype, risks, medical management.

The phenotype of the disorder, its treatment and preventive options need to be discussed. With respect to treatment and prevention, it should be discussed whether these procedures are experimental or not and what the proven or suspected advantages and disadvantages are. The risks for the different phenotypical features need to be presented, preferably as cumulative risks against the background of normal cumulative population risk (*e.g.* 20 % remaining risk to develop breast cancer), rather than as relative risks (*e.g.* twofold risk to develop breast cancer). Since risk perception, risk recollection, and perception of the benefits of medical surveillance and prophylactic surgery may be inaccurate[11-20], written material should be provided which summarizes all important issues discussed during counselling.

Inheritance and reproductive options

Families may have strong misconceptions of Mendelian inheritance and the basic statistics involved. These misconceptions, sometimes referred to as 'family mathematics', may seriously interfere with rational decision making with respect to *e.g.* participation in DNA testing or medical surveillance programs. Bearing this in mind, the general mechanism of gene transmission and the probability of carrying, respectively transmitting the disease gene need to be discussed including the options for DNA testing if these are available (see also the paragraph on psychological aspects). As in the counselling on other hereditary disorders,

information on the various reproductive options should be provided. Although technically the option for prenatal DNA testing is available for a number of human cancer syndromes, there appears to be little demand for it, presumably because of the late-onset and (potentially) treatable nature of the most frequent human cancer syndromes.

Psychological aspects

Members of families with human cancer syndromes may carry various psychological burdens, including those associated with the early death of parents and other close relatives, the fear of losing other relatives (including children) and the fear to develop cancer themselves. Psychological issues need to be addressed during counselling, because this may help to better individualize the counselling and may also signal possible reasons for referral for further psychological support. Also, severe psychological distress is a contra-indication for predictive DNA testing. Among the psychological aspects which need to be discussed is the fact that, contrary to most medical tests, the outcome of genetic analysis is not only relevant to the consultand, but may also have significant personal consequences for other family members (who may not even be aware that such an analysis is being carried out in their family). Ways to distribute information on the genetic risks and medical management options to relatives, as well as the risks of acting as the 'messenger' of potentially threatening information in the family, should be addressed.

Of special concern are the psychological aspects of predictive (*i.e.* presymptomatic) DNA testing, which have been the subject of most of the psychological research in hereditary cancer. Most of these studies have been performed on individuals participating in research-based DNA testing which followed careful pre and post testing counselling protocols. It appears that at least short- and middle-term psychological distress in these individuals is relatively small[21] and that this distress depends more on pretest expectations, mood and social support than on the results of the test itself[22,23].

Ongoing research now tries to identify risk factors for adverse psychological reactions in order to offer better psychological support during genetic counselling and after. Because of the potential risk for significant adverse personal reactions and family disruption[21,24], potential disadvantages of predictive testing should be discussed with the consultands as part of an informed consent procedure.

Genetic discrimination

Information on the genetic status of individuals with respect to hereditary cancer predisposition might be used by employers and insurance companies. As a result, it could be more difficult for individuals with such a predisposition to become or remain employed, or to obtain health, disability or death insurance coverage at normal rates (or at any rate at all)[25-29]. The practical aspects of these issues must be discussed in the genetic counselling as part of the informed consent procedure of predictive and diagnostic DNA testing. Each country has its own rules and legislation with respect to genetic discrimination (*those of the Netherlands are summarized in the section 4.3.1 of chapter 4*).

Registration

As one of the final steps in genetic counselling, relatives from families with human cancer syndromes may be offered inclusion in a hereditary cancer registry, in order to enhance participation in medical surveillance and facilitate research on those syndromes. *Hereditary cancer registries are further discussed in chapter 2, section 2.4*

Organizational aspects:

To cope with the growing demand for genetic diagnosis and counselling of cancer patients and their families, a number of hospitals and universities have organized 'cancer family clinics' which accommodate a team of clinical geneticists, genetic counsellors/nurses/associates, often psychologists/medical social workers, usually backed-up by a DNA and cytogenetics laboratory with its own staff, and either include medical oncological specialists (*e.g.* surgeons, gynaecologists, medical oncologists) or work in close collaboration with those specialists. These clinics may function as a part of normal health care (as *e.g.* in the Netherlands) or may operate in a research setting.

Depending on the outcome of the genetic diagnosis and counselling, consultands and relatives may need to be referred (back) for medical surveillance and/or preventive surgery. To enhance the participation in medical surveillance and to determine the effect of the preventive options, families may be offered inclusion in hereditary cancer registries

A flow-chart of the above mentioned processes in our own Department is presented in figure 1. Not all of the steps summarized in this figure are available or appropriate in all diagnostic and counselling situations.

The organizational aspects and the use of hereditary cancer registries are further discussed in the methods sections of chapters 3,4 and 2, respectively.

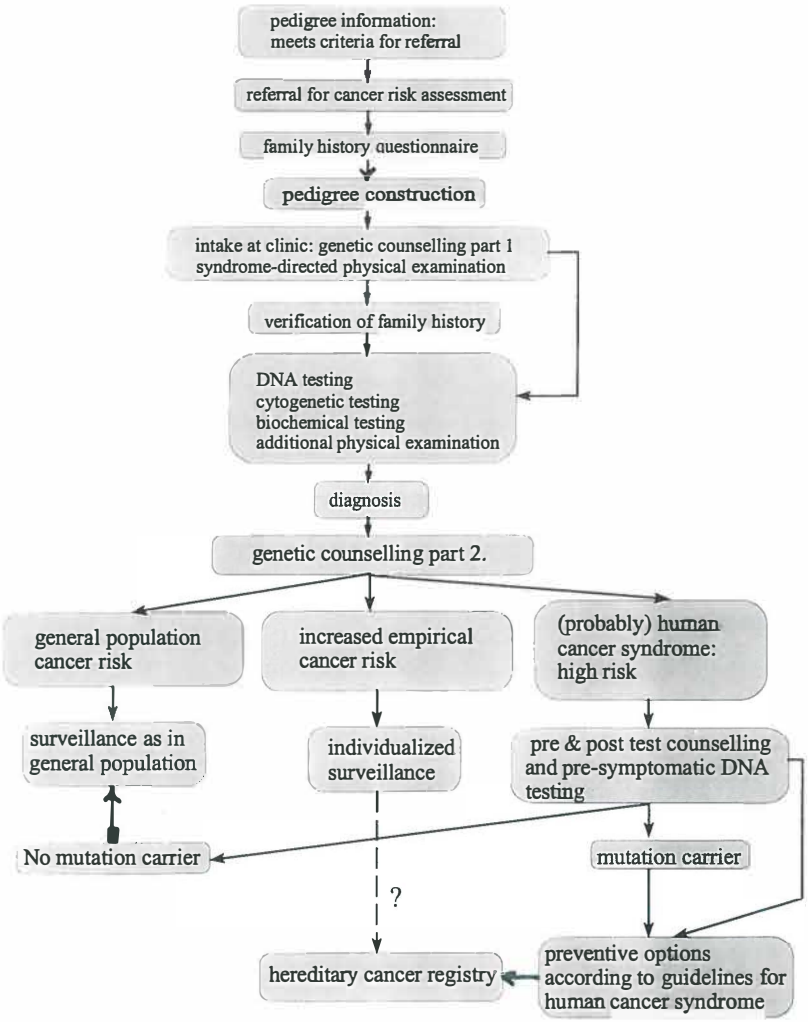

***Figure 1**. Flowchart of genetic diagnosis and counselling for cancer and follow-up. Not all of the steps summarized in this figure are available or appropriate in all diagnostic and counselling situations. ? = registration of the outcome of screening in this group is important, but it is not a classical task of the hereditary cancer registries*

1.3 Human cancer syndromes

1.3.1 Epidemiology

Human cancer syndromes account for an estimated 1-10 % of the total cancer burden. Depending on the type of tumour involved, the estimates of the hereditary proportion of cases range from less than 5 %, *e.g.* in the case of renal cancer[30,31] to 5-10 %, *e.g.* in the case of breast cancer[32,33] and sometimes even more, *e.g.* approximately 25 % in the case of medullary thyroid carcinoma[34] and 40 % in the case of retinoblastoma[35]. These estimates do not stem from a complete molecular understanding of the molecular basis of the hereditary predisposition for these tumours, instead, they have usually been based on the observed familial clustering of the different types of cancer[32], and in some cases on segregation analysis trying to fit the observed familial and non-familial cases to genetic models[36,37]. Given the fact that neither the presence nor absence of familial clustering of cancer is proof of the hereditary nature, these estimates should at best be considered imprecise. Looking at the proportion of cancer patients with linkage to particular gene loci, with proven germline mutations and/or clinically diagnosed with human cancer syndromes will allow us to estimate the minimal proportion of hereditary cases.

1.3.2 Oncogenesis

Cancer results from the accumulation of genetic damage, a multi-step process of mutation of genes essential for normal growth control, which promotes clonal selection of cells with increasingly aggressive behaviour[38]. This process is referred to as oncogenesis. The genes involved in oncogenesis have been classified in the past as either '*tumour suppressor genes*', requiring the mutation of both copies to initiate tumour development or *proto-oncogenes*, requiring the mutation of only one copy for the initiation. Kinzler and Vogelstein have reclassified these genes as either '*gatekeepers*', which directly control cellular proliferation, and '*caretakers*', which maintain genetic stability[39]. Both classifications are illustrated in figure 2

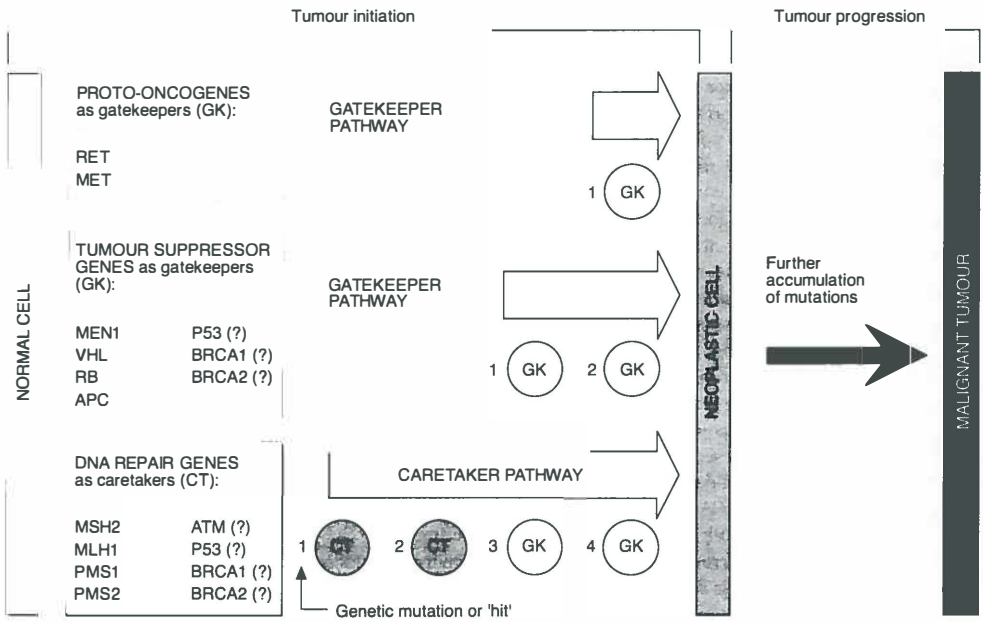

Figure 2. Schematic representation of the different pathways to tumour initiation, involving gatekeeper (GK) and caretaker (CT) genes (modified from Kinzler & Vogelstein[39]). In the pathway in which a proto-oncogene acts as a gatekeeper (top), only one mutation is required to initiate oncogenesis. In contrast, if a tumour suppressor gene functions as gatekeeper (middle), mutations of both gatekeeper alleles are required. Finally, in the caretaker pathway (bottom) a total of four mutations, resulting in the inactivation of two alleles of a caretaker gene (causing genetic instability) and two alleles of a gatekeeper gene (leading to loss of growth control), are necessary to drive oncogenesis. From: Pearson PL and van der Luijt RB. The genetic analysis of cancer. J Int Med 1998;243:413-417 (used with permission).

Most of the mutations in oncogenesis arise spontaneously in the somatic cells during DNA replication or are caused by mutagenic agents (*e.g.* ionizing radiation and carcinogens). These mutations are referred to as 'somatic mutations' and occur in genes which were inherited as normal ('wild-type') copies from the parents. A smaller proportion of mutations is inherited from the parents and these mutations are referred to as 'germline mutations'. Contrary to somatic mutations, they are present in all cells of an individual. This highly increases the probability that the total number of mutations needed for cancer development will be reached. For some of the tumour types in some human cancer syndromes this life-time risk is 90-100 % (*e.g.* the risk of retinoblastoma in hereditary retinoblastoma, or medullary thyroid cancer in multiple endocrine neoplasia type 2), in other cases it is lower (*e.g.* 70-90 % risk of colorectal cancer and 30-50 % risk of endometrial cancer in hereditary non-polyposis colorectal cancer (HNPCC)). To illustrate the genetic pathways of cancer development as part of the human cancer syndromes, three examples are presented below.

The process of retinoblastoma development comes probably closest to the most simple multi-step model known in oncogenesis: the '2-hit model' of Knudson[40], who postulated on the grounds of age-incidence curves of unilateral compared with bilateral retinoblastoma that 2 independent gene mutations *(i.e.* 'hits') were necessary for that tumour to develop. Comings[41] speculated that these two gene mutations involved the two copies of one and the same gene (one copy on each of a pair of chromosomes). Subsequently, these mutations were identified as mutations of the gatekeeper and tumour suppressor retinoblastoma gene (RB1) on the long arm of chromosome 13. In hereditary retinoblastoma, one of the mutations has been inherited from one of the parents and because all retina cells carry this mutation, the chance that in one of these cells a second hit occurs and subsequently a retinoblastoma develops, is high (average of 90 %).

In multiple endocrine neoplasia, type 2 (MEN2), a germline mutation is present in the gatekeeper proto-oncogene RET, leading to uncontrolled cell growth stimulating signalling. As a result of this stimulation, tumours develop. *MEN2 is further discussed in appendices I and II.*

In HNPCC, germline mutations can be demonstrated in caretaker genes known as DNA mismatch repair (MMR) genes. These genes are responsible for the repair of a particular type of DNA damage, referred to as 'mismatches'. Contrary to the genes of hereditary retinoblastoma and MEN2, which are placed directly in the oncogenetic pathway, germline mutations in the MMR genes have an indirect effect. Once, in the course of life, a somatic mutation hits the normal ('wild type')

counterpart of the MMR gene mutated in the germline, DNA mismatch repair becomes deficient. As a direct result of this, mismatch type of mutations accumulate in a range of genes which are particular susceptible to the occurrence of those mutations, namely those which contain many repetitive simple DNA sequences. Some of these genes are important to maintain normal growth control. When they get mutated, cancer develops. *HNPCC is further discussed in appendices III and IV.*

1.3.3 Tumour spectrum, tumour risks and genotype-phenotype associations

No human cancer syndrome presents with an increased risk for all types of tumours. Instead, each of these syndromes has its own specific tumour spectrum. In addition to the oncological manifestations, non-oncological features may occur as well, *e.g.* craniofacial dysmorphisms and skeletal anomalies. *The phenotypes of the human cancer syndromes are reviewed in chapter 2 and are more fully presented in the Familial Cancer Database, which is the subject of chapter 5.* Phenotypes of the human cancer syndromes are being established by two different strategies. The most widely used approach is the collection of clinical observations from affected families and the statistical analysis of these. *This approach is demonstrated in appendix III.*

If features are only rarely observed, then the statistical approach may not be able to determine whether or not those features are indeed part of the phenotype. Recent scientific advances allow sometimes for a second approach: the study of a particular clinical feature for molecular characteristics that are typical for the human cancer syndrome involved.

This approach is demonstrated in appendix IV for a malignant fibrous histiocytoma occurring in an HNPCC patient and is discussed for Hirschsprung disease in MEN2A in appendix II.

Crucial to the medical management and genetic counselling of the human cancer syndromes are the specific tumour risks associated with those syndromes. Very different risks are usually associated with the different tumours from the tumour spectrum. These differences may theoretically be explained by assuming differences between the target tissues with respect to:

- exposure to mutagenic agents (which may cause somatic mutations),
- rate of stem cell proliferation (which influences the rate of accumulation of somatic mutations),
- expression of the cancer syndrome gene (reflecting its physiological role in that tissue)
- the contribution of the mutant cancer syndrome gene to the tissue-specific oncogenetic pathway

For each tumour, the risk of developing it is established by the statistical analysis of observations in affected families, taking into account the occurrence of tumours in the general population. *This method is demonstrated in appendix III for urinary tract cancer in HNPCC.*

The phenotype of a particular human cancer syndrome may depend on characteristics of the associated germline mutation. A classical example is MEN2, where depending on the type of mutation in the RET gene, patients present with the aggressive phenotype of MEN2B, or the milder phenotypes of MEN2A or familial medullary thyroid cancer. *Some MEN2A mutations are also associated with Hirschsprung disease, discussed in appendix II.* Other examples are the association between the number of adenomatous polyps and extra-gastrointestinal manifestations and the type of APC gene mutation in familial adenomatous polyposis (FAP)[42], and the association of phaeochromocytomas in von Hippel Lindau disease with particular VHL gene mutations[43].

1.3.4 Treatment and preventive options

The treatment of cancer occurring as part of a human cancer syndrome may need to differ from treatment of non-hereditary cancer with a similar clinical stage/grade. This difference is caused by the fact that there is an increased risk to develop multiple primary tumours, warranting more extended surgery, or because tissue response to chemo- or radiation therapy may be abnormal. Practical examples of such differences are the subtotal colectomy (rather than more limited resection) for HNPCC associated colon cancer[44], and the dose reduction of radiotherapy for

lymphomas in ataxia telangiectasia patients (who have an increased radiosensitivity)[45]. For a number of human cancer syndromes, preventive options, ranging from periodic medical surveillance to prophylactic surgery and chemoprevention have been formulated. *These preventive options have been reviewed in chapter 2.*

1.4 References

1 Murphy EA, Chase GA. *Principles of Genetic Counseling*. Chicago: Year Book Medical Publishers, 1975;86

2 Spraycar M. *Stedman's Medical Dictionary*. Baltimore: Williams & Wilkins, 1995;387

3 Harper PS. *Practical Genetic Counselling*. Oxford: Butterworth-Heinemann, 1998;6

4 Peters JA, Biesecker BB. Genetic counseling and hereditary cancer. *Cancer* 1997;**80**:576-586.

5 d'Errico A, Taioli E, Chen X, Vineis P. Genetic metabolic polymorphisms and the risk of cancer: A review of the literature. *Biomarkers* 1996;**1**:149-173.

6 Perera FP. Molecular epidemiology: Insights into cancer susceptibility, risk assessment, and prevention. *J Natl Cancer Inst* 1996;**88**:496-509.

7 Bartsch H, Hietanen E. The role of individual susceptibility in cancer burden related to environmental exposure. *Environ Health Perspect* 1996;**104(suppl.3)**:569-577.

8 Perera FP. Environment and cancer: Who are susceptible? *Science* 1997;**278**:1068-1073.

9 Aarnio M, Mecklin JP, Aaltonen LA, Nystrom-Lahti M, Jarvinen HJ. Life-time risk of different cancers in hereditary non- polyposis colorectal cancer (HNPCC) syndrome. *Int J Cancer* 1995;**64**:430-433.

10 Peters JA, Stopfer JE. Role of the genetic counselor in familial cancer. *Oncology* 1996;**10**:159-166.

11 Evans DGR, Blair V, Greenhalgh R, Hopwood P, Howell A. The impact of genetic counselling on risk perception in women with a family history of breast cancer. *Br J Cancer* 1994;**70**:934-938.

12 Evans DGR, Burnell LD, Hopwood P, Howell A. Perception of risk in women with a family history of breast cancer. *Br J Cancer* 1993;**67**:612-614.

13 Vernon SW, Vogel VG, Halabi S, Bondy ML. Factors associated with perceived risk of breast cancer among women attending a screening program. *Breast Cancer Res Treat* 1993;**28**:137-144.

14 Black WC, Nease RF, Tosteson ANA. Perceptions of breast cancer risk and screening effectiveness in women younger than 50 years of age. *J Natl Cancer Inst* 1995;**87**:720-731.

15 Lerman C, Lustbader E, Rimer B, et al. Effects of individualized breast cancer risk counseling: A randomized trial. *J Natl Cancer Inst* 1995;**87**:286-292.

16 Smith BL, Gadd MA, Lawler C, et al. Perception of breast cancer risk among women in breast center and primary care settings: Correlation with age and family history of breast cancer. *Surgery* 1996;**120**:297-303.

17 Hughes C, Lerman C, Lustbader E. Ethnic differences in risk perception among women at increased risk for breast cancer. *Breast Cancer Res Treat* 1996;**40**:25-35.

18 Watson M, Duvivier V, Wade Walsh M, et al. Family history of breast cancer: what do women understand and recall about their genetic risk? *J Med Genet* 1998;**35**:731-738.

19 Iglehart JD, Miron A, Rimer BK, Winer EP, Berry D, Shildkraut JM. Overestimation of hereditary breast cancer risk. *Ann Surg* 1998;**228**:375-383.

20 Watson M, Lloyd S, Davidson J, et al. The impact of genetic counselling on risk perception and mental health in women with a family history of breast cancer. *Brit J Cancer* 1999;**79**:868-874.

21 Croyle RT, Achilles JS, Lerman C. Psychologic aspects of cancer genetic testing: A research update for clinicians. *Cancer* 1997;**80**:569-575.

22 Marteau TM, Croyle RT. The new genetics: Psychological responses to genetic testing. *Br Med J* 1998;**316**:693-696.

23 Dudok de Wit AC, Tibben A, Duivenvoorden HJ, Niermeijer MF, Passchier J. Predicting adaptation to presymptomatic DNA testing for late onset disorders: who will experience distress? *J Med Genet* 1998;**35**:745-754.

24 Lerman C, Croyle RT. Emotional and behavioral responses to genetic testing for susceptibility to cancer. *Oncology* 1996;**10**:191-199.

25 Harper PS. Insurance and genetic testing. *Lancet* 1993;**341**:224-227.

26 Culliton BJ. Genes and discrimination. *Nat Med* 1995;**5**:385

27 Hudson KL, Rothenberg KH, Andrews LB, Kahn MJE, Collins FS. Genetic discrimination and health insurance: an urgent need to reform. *Science* 1995;**270**:391-393.

28 Parens E. Glad and terrified: On the ethics of BRCA1 and 2 testing. *Cancer Invest* 1996;**14**:405-411.

29 Lapham EV, Kozma C, Weiss JO. Genetic discrimination: perspectives of consumers. *Science* 1996;**274**:621-624.

30 Griffin JP, Hughes GV, Peeling WB. A survey of the familial incidence of adenocarcinoma of the kidney. *Br J Urol* 1967;**39**:63-66.

31 Mclaughlin JK, Mandel JS, Blot WJ, Schuman LM, Mehl ES, Fraumeni JF, Jr. A population--based case--control study of renal cell carcinoma. *J Natl Cancer Inst* 1984;**72**:275-284.

32 Lynch HT, Lynch JF. Breast cancer genetics in an oncology clinic: 328 consecutive patients. *Cancer Genet Cytogenet* 1986;**22**:369-371.

33 Claus EB, Schildkraut JM, Thompson WD, Risch NJ. The genetic attributable risk of breast and ovarian cancer. *Cancer* 1996;**77**:2318-2324.

34 Saad MF, Ordonez NG, Rashid RK, et al. Medullary carcinoma of the thyroid. A study of the clinical features and prognostic factors in 161 patients. *Medicine Baltimore* 1984;**63**:319-342.

35 Vogel F. Genetics of retinoblastoma. *Hum Genet* 1979;**52**:1-54.

36 Newman B, Austin MA, Lee M, King MC. Inheritance of human breast cancer: evidence for autosomal dominant transmission in high-risk families. *Proc Natl Acad Sci USA* 1988;**85**:3044-3048.

37 Carter BS, Beaty TH, Steinberg GD, Childs B, Walsh PC. Mendelian inheritance of familial prostate cancer. *Proc Natl Acad Sci USA* 1992;**89**:3367-3371.

38 Fearon ER. Human cancer syndromes: Clues to the origin and nature of cancer. *Science*1997;**278**:1043-1050.

39 Kinzler KW, Vogelstein B. Cancer-susceptibility genes - Gatekeepers and caretakers. *Nature* 1997;**386**:761-763.

40 Knudson AG, Jr. Mutation and cancer: statistical study of retinoblastoma. *Proc Natl Acad Sci U S A* 1971;**68**:820-823.

41 Comings DE. A general theory of carcinogenesis. *Proc Natl Acad Sci U S A* 1973;**70**:3324-3328.

42 Fodde R, Meera Khan P. Genotype-Phenotype correlations at the adenomatous polyposis coli (APC) gene. *Crit Rev Oncogenesis* 1997;**6**:291-303.

43 Chen F, Kishida T, Yao M, et al. Germline mutations in the von Hippel-Lindau disease tumor suppressor gene: Correlations with phenotype. *Hum Mutat* 1995;**5**:66-75.

44 Lynch HT, Smyrk T. Hereditary nonpolyposis colorectal cancer (Lynch syndrome) - An updated review. *Cancer* 1996;**78**:1149-1167.

45 Abadir R, Hakami N. Ataxia telangiectasia with cancer. An indication for reduced radiotherapy and chemotherapy doses. *Br J Radiol* 1983;**56**:343-345.

Chapter 2
Familial and Hereditary cancer: review of risks, differential diagnosis and preventive options.

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2.1 Introduction

Cancer results from an accumulation of mutations in genes which are important for maintaining normal cell growth and for programmed cell death. In most cases these mutations arise in the course of life, spontaneously or through the action of mutagenic agents (e.g. ionizing radiation and carcinogens). These mutations are referred to as 'somatic' mutations and the cancer they cause as 'non-hereditary'. In an unknown, but possibly significant number of cases, susceptibility to carcinogens is influenced by the inherited variation in the action of enzymes responsible for (pro)carcinogen metabolism. The hereditary variation of these enzymes generally moderately influences cancer risk and may contribute to an unknown proportion of the familial clustering of cancer.

In contrast, some individuals carry a germline mutation of a gene which confers a high risk of developing cancer, often at a relatively early age[1,2]. These malignancies are referred to as *hereditary cancer* and contribute an estimated 1%-10% to the total cancer burden. This proportion strongly depends on the tumour type. For example, an estimated 5-10 % of breast or ovarian cancer cases are hereditary[3,4], whereas less than 1-2.5 % of renal cancer is [5,6]. Hereditary cancer is the main or only feature of a group of hereditary disorders known as the *hereditary cancer syndromes, human cancer syndromes* or *hereditary cancer predisposition syndromes*. The term *familial cancer* is used by some as a synonym of hereditary cancer, but is more commonly defined as simply the familial occurrence of cancer, not necessarily due to genetic predisposition. In addition to the human cancer syndromes, there are a number of inherited disorders which are associated with a more moderate increase of cancer risk.

The general public shows a growing interest in the hereditary aspects of cancer and increasingly confronts their physicians with questions on cancer risk related to their family histories. Physicians, on their part, are interested, particularly because recognizing inherited cancer predisposition may help them target tumour prevention programs. Also, treatment of hereditary cancer may differ from that in its non-hereditary counterparts. Examples are the preference of subtotal colectomy instead of more regional resection in case of colon cancer found in hereditary non-polyposis colorectal Cancer (HNPCC, Lynch syndrome) and the dose reduction in the radiotherapy of lymphomas in patients with ataxia telangiectasia. Notably, consequences in terms of family risk and psychosocial impact are different and it is therefore of importance to correctly identify hereditary and non-hereditary cancer.

In this paper we want to provide the clinician with a review of familial clustering of cancer and of cancer as part of inherited disorders. This review is focused on presenting data which are of practical use when considering the hereditary nature of cancer in a patient and his or her family and in answering patients' questions on cancer risk related to their personal or family history, possibly interacting with other personal risk factors. We first discuss recognition of hereditary cancer in general (section 2.2) and after that focus on the clinical genetic differential diagnosis of the fifteen most frequent types of solid cancer diagnosed in Western-Europe and North-America; on the family history as a risk factor for these types of cancer; and on the current options for the early detection and prevention of these tumours (section 2.3). We also review the role of hereditary cancer registries in the prevention of hereditary cancer (section 2.4). Although we acknowledge the obvious importance of the therapy of hereditary cancer and the ethical, psychosocial, legal and economical issues involved in hereditary cancer predisposition, these aspects are beyond the scope of this paper.

2.2 Recognition of hereditary cancer

In some individuals with a human cancer syndrome, clinical features are very striking if not pathognomonic, thereby facilitating the diagnosis. Examples are the typical facial features, palmoplantar pitting, skeletal abnormalities and multiple basal cell carcinomas in the basal cell carcinoma (Gorlin) syndrome[7,8]. The presence of Marfanoid habitus, oral neuromas and medullary thyroid cancer leads to the diagnosis of multiple endocrine neoplasia type 2B[9] and the detection of hundreds of adenomatous intestinal polyps to the diagnosis of familial adenomatous polyposis[10]. However, in many other human cancer syndromes one needs to look for additional diagnostic clues since the clinical features in one patient alone are often not typical enough to diagnose those syndromes.

2.2.1 The family history of cancer.

A family history of cancer may provide important clues to the diagnosis hereditary cancer. However, familial occurrence of cancer is not necessarily due to a strong genetic predisposition. Because cancer is a very common disease, chance alone may cause clustering in these families. In addition to chance, familial clustering of cancer might also be due to shared environmental risk factors or combinations of both hereditary risk factors and environmental factors[11-13]

A large number of known human cancer syndromes have an autosomal dominant inheritance, in which there is a 50 % risk for children of an affected parent to inherit the mutant gene. Depending on the penetrance of that mutant gene, up to 100 % of the carriers of that mutation will develop the disease. The presence of affected relatives in successive generations, in particular affected children having affected parents, should therefore alert the physician to the possibility of hereditary cancer. However, there are several reasons why hereditary cancer need not always manifest itself in this striking pedigree pattern. Apart from the fact that the family history may be incomplete or inaccurate[14], there is the possibility of non-penetrance, meaning that an individual carrying the mutant gene in question does not show any clinical features of the disease. Many of the genes involved have an age-dependent penetrance resulting in an increasing chance of having developed the disease with increasing age, but in many cases never reaching 100 %. In addition, clinical expression of a mutant gene may be sex-specific as in hereditary (breast -) ovarian cancer and in hereditary prostate cancer.

Unclear pedigree patterns may also be caused by *de novo* mutations, meaning that the parents of a patient affected with autosomal dominant cancer do not themselves carry the mutant genes but transmitted the mutation to their child because of a spontaneous mutation in their germ cells. Well known examples of this mechanism are familial adenomatous polyposis (25-30 % of cases de novo[10]) and multiple endocrine neoplasia type 2B (50 % of cases de novo[15]).

The absence of a dominant pattern in hereditary cancer may of course be caused by a different type of inheritance. Hereditary cancer may also be inherited as an autosomal recessive trait, typically manifesting itself in a pedigree as unaffected parents having affected children. There is a 25 % risk for a child of both parents carrying the mutant gene to be affected.

Well known examples are Fanconi anaemia, Bloom syndrome, xeroderma pigmentosum (XP) and ataxia telangiectasia (AT). Matters can be complex since *e.g.* in female carriers of one copy of the mutant AT gene, although AT does not develop, the risk of breast cancer is increased[16,17].

X-linked inheritance is characterized by a 50 % risk for a son of a mother carrying the mutation to be affected. The risk and disease severity for daughters depend on the disease being X-linked recessive or dominant. No male-to-male transmission is seen and all daughters of affected males inherit the mutation. X-linked inheritance is seen infrequently in hereditary cancer, probably because X-linked inherited disorders generally severely reduce the reproductive fitness of the affected males. X-linked lymphoproliferative syndrome is an example of an X-linked condition with frequent oncological complications[18].

With regard to the family history, we can conclude that a positive family history of cancer fitting one of the known inheritance patterns may suggest a hereditary predisposition to cancer (depending on details, more or less strongly), but in itself is no proof. *Vice versa*, the absence of a family history of cancer in a cancer patient does not exclude the presence of an inherited strong cancer predisposition given the possibilities of non-penetrance, age-dependent penetrance, de novo mutation, autosomal recessive inheritance (and a small number of siblings), and X-linked inheritance (and small number of brothers, maternal uncles etc.).

2.2.2 Additional characteristics

Finding additional features of hereditary cancer in a cancer patient and or his or her family supports the diagnosis:

- Early age at diagnosis of a tumour as compared to the distribution of the age at diagnosis in the general population is an important characteristic. For example, in Hereditary Non-Polyposis Colorectal Cancer (HNPCC, Lynch syndrome), most colorectal cancers occur before the age of 50 years[19], while the opposite is true in the general population. In families with Hereditary Breast-Ovarian Cancer female carriers of the mutant BRCA1-gene have a risk of 50 % of developing breast cancer before the age of 50[20], as compared with a risk of 2 % in the general female population[21].

- Familial clustering of tumours in the absence of known important environmental risk factors may also be a clue to a strong genetic predisposition, *e.g.* familial lung cancer in non-smokers.

- Multiple primary tumours at the same site, bilateral, or in different organ types should alert to the possibility of a hereditary cancer predisposition. Certain combinations of different cancer types in a patient, but also in a family as a whole, are specifically associated with human cancer syndromes, *e.g.* colorectal and endometrial cancer with HNPCC[22], breast, ovarian and prostate cancer with

hereditary breast-ovarian cancer[23], breast cancer and sarcomas with Li-Fraumeni syndrome[24] and medullary thyroid cancer and phaeochromocytoma with multiple endocrine neoplasia type 2[25].

- The presence of multiple precursor lesions may be another feature, for example the polyps in the different polyposis syndromes (see section 2.3.4).

- Congenital malformations are additional characteristics of some of the human cancer syndromes, *e.g.* the macrocephaly in Cowden syndrome, the omphalocele in Beckwith-Wiedemann syndrome and the ambiguous genitals in Denys-Drash syndrome (see table 1).

- the development of cancer in an unusual gender context, particularly breast cancer in males, which is associated with germline BRCA2, and sometimes BRCA1, gene mutations (see section 2.3.2).

- The known presence of a hereditary disorder in a relative of the cancer patient should prompt reevaluation of the patient in this context. Possibly his or her tumour could be interpreted as a complication of that hereditary disorder.

In summary, unusual features, in terms mentioned above, in the cancer patients and/or their families which should alert the physician to the possibility of an inherited disorder with cancer predisposition. In a growing number of cases, DNA testing will be of help in making a definite diagnosis, which would also allow for presymptomatic DNA testing in relatives. In some syndromes chromosomal, biochemical or haematological tests can be used to confirm or reject the clinical suspicion of those disorders, *e.g.* the specific tests for spontaneous and induced chromosomal breakage and persistent increased presence of fetal type haemoglobin (HbF) in Fanconi anaemia. In other hereditary disorders, the diagnosis can be made if certain characteristic physical findings (*e.g.* specific congenital abnormalities) are present and/or the family history meets certain criteria, *e.g.* finding the combination of optic glioma, sphenoid dysplasia, 6 large cafe au lait spots and 2 Lisch noduli of the iris in a patient, would be more than enough to diagnose neurofibromatosis type 1.

2.3 Familial risks, clinical genetic differential diagnosis and preventive options

The following sections of this article will discuss in alphabetical order the most frequent types of cancer found in North-American and Western European men and women [26].

First, we review the risks to develop a specific type of cancer associated with a family history of that type or other types, and, if available, prognostic and histological features of those familial tumours. These data may be particularly useful in answering questions raised by patients with a moderate family history of cancer. Risks are presented in the literature as either cumulative (life-time) risks, or risks relative to that in the general or a specific control population, the relative risks (RR)[27]. It should be kept in mind that most risk estimates provided in the literature restrict family history to first-degree relatives only. Therefore, these estimates may be (far) too low for individuals with a strong family history of cancer and may be too high for individuals from large families with few affected relatives. In addition, most studies have been performed in North-American and Western-European populations and the outcome of these studies might not apply to populations with a different ethnic/genetic background.

Second, we review the hereditary disorders presenting with the common types of cancer and also less common malignant or benign tumours, occurring at the same anatomical sites if these are relevant from a hereditary point of view. In rare human cancer syndromes it may be difficult to decide which tumours should be regarded as a true feature of the disease and which should not, because a proportion of the tumours reported in the various patients and families with these disorders will represent coincidental findings. For this review, we have limited ourselves to the more prevalent features of the human cancer syndromes in order to avoid introducing doubtful associations. Still, this also means that some were probably left out which will ultimately be proven to belong to the syndromes' phenotypes.

Falling outside the scope of this review is the wide range of hereditary variants of genes coding for enzymes involved in carcinogen metabolism, which are generally associated with moderate cancer risks, because testing for these variants is not (yet) part of the common genetic analysis of (familial) cancer cases.

Third and last, we review the preventive options currently available to individuals at high genetic risk for developing the selected types of cancer, as well as the preventive value of maintaining hereditary cancer registries.

For a summary of the hereditary disorders mentioned in the following paragraphs, the reader is referred to table 1.

2.3.1 Bladder

Relative risks

Familial clustering of bladder cancer has been reported several times [28-32] and occurs in approximately 6 % of cases[33]. Kantor *et al.* [33] demonstrated in a study of 2,982 bladder cancer patients and 5,782 controls, that having an affected first-degree relative increases the risk of bladder cancer. Relative risks (RR's) vary dependent of the age at diagnosis of the tumour: 2.7 below age 45, 1.7 for ages 45-64 and 1.3 for the age of 65 and older. Cigarette smoking in the setting of a family history of bladder cancer was shown by the authors to strongly increase the risk of developing this tumour. Relative risks of up to 28.1 in heavy smokers (60 + cigarettes/day) were calculated as compared with an RR of 2.1 in heavy smokers with a negative family history. Although the risks appear to be highest for smokers and the advice to stop smoking may have an important effect on reducing cancer risk, non-smokers with a positive family history are still at a somewhat increased risk (RR of 1.5-1.8), as demonstrated by Kantor *et al.*[33] and Kramer *et al.*[34].

Bladder cancer and hereditary disorders

As yet, bladder cancer is not considered an established part of any known human cancer syndrome or other hereditary disorders.

Preventive options

Screening for bladder cancer is discussed as part of screening for urinary tract cancer in general in section 2.3.6 (Kidney). In patients with a family history of bladder cancer it might be important to try to reduce exposure to known environmental risk factors for this tumour, including smoking. Experiments in mice have hinted at a future option of chemoprevention of bladder cancer, possibly through use of ketoprofen or sulindac [35]. As yet, its value for use in humans is unknown.

2.3.2 Breast

Relative risks

Given the high risk of breast cancer in women[36], familial occurrence of breast cancer is not rare. A family history of breast cancer has been reported in 14% to 30% of female breast cancer patients[3,37]. In contrast, familial male breast cancer is very rare[38-40]. Having a relative with breast cancer is one of the strongest breast cancer risk factors. Pooled estimates of relative risks for women with an affected relative are, depending on the type of relative with breast cancer, 1.9 (any type of affected relative), 2.1 (first-degree relative), 2.0 (mother), 2.3 (sister), 1.8 (daughter), 3.6 (mother and sister) and 1.5 (second degree)[41]. In general, the risk of breast cancer in relatives does not appear to depend heavily on the gender of their affected family member[42-45]. Depending on the number of affected relatives, the degree of kinship and the age at onset of the tumours, the empiric risk of developing breast cancer for a female relative may be close to 50 % (even higher if she already has children with breast cancer). Compared with the general population, breast cancer has also been observed more frequently in women with a first-degree relative with ovarian cancer (RR 6.7, cumulative risk 14-42 %)[46-48], endometrial cancer (RR 3.9, cumulative risks 34 %)[46,47], colorectal cancer (RR 1.2-2.0)[47,49] and prostate cancer (RR 4.7, cumulative risk 33 %)[46,47].

Life-time risks

Several models have been developed to estimate breast cancer risk given a specific family situation[50]. The model by Gail *et al.* [51-55], focuses on white females who are being examined annually and takes into account the risk factors of first-degree affected relatives, age at menarche, age at first live birth and number of previous biopsies. The model of Claus *et al.*[48,56,57] is the one most widely used in cancer family clinics, as it offers figures for a wider range of family history types (first as well as second-degree affected relatives)(table 2). Users of the Gail and Claus models should be aware of their specific limitations[50,58]. For example, the risk tables may need to be adapted for use in different countries and or ethnic groups to account for differences in genetic background and exogenous risk factors[59]. Also, the models do not take into account the number and ages of unaffected relatives and therefore overestimate cancer risk in families with many healthy relatives[60]. On the other hand, the models may underestimate cancer risk for women with family histories strongly suggestive of hereditary breast(-ovarian) cancer (*e.g.* for those with an affected mother and daughter).

Table 2 **Cumulative life-time risk (%) of developing breast cancer in women with a family history of breast cancer (modified after Claus *et al.*[57]).**

	Degree of kinship with affected relative and number of affected relatives							
	one 1st degree	two 1st degree						one 2nd degree
Age at diagnosis of relative diagnosed at youngest age:		Age at diagnosis of second 1st degree relative						
		20-29	30-39	40-49	50-59	60-69	70-79	
20-29	21.1	48.4	46.0	43.4	39.7	35.4	30.8	14.2
30-39	16.5		43.7	39.9	35.3	30.2	25.2	12.0
40-49	13.2			35.4	30.0	24.6	20.0	10.4
50-59	11.0				24.5	19.5	15.8	9.4
60-69	9.6					15.6	12.8	9.4
70-79	8.8						10.9	8.3

To illustrate how the number and degree of affected relatives may affect breast cancer risk, this table lists cumulative risks to develop breast cancer depending on the number of first and second-degree affected relatives and the age at diagnosis in these relatives. For example, a woman with 1 first-degree relative, who was diagnosed with breast cancer at the age of 33 years, has a cumulative risk to develop breast cancer of 16.5 %. If that same woman had another affected first-degree relative, diagnosed at the age of 43 years, her risk would be 39.9 %. If she only had 1 second-degree relative aged 68 years at the time of diagnosis, her risk would be 9.4 %. The original tables published by Claus et al.[57] allow calculations for a wider and more detailed range of family histories and allow for correction for past disease-free years of the individual for whom risks are calculated.
(modified from: Oosterwijk JC, Sijmons RH, Menko FH, Chorus AMJ, Rookus MA. Ned Tijdschr Geneesk 1995;139:423-428)

Interaction of the family history with other risk factors

Given a positive or negative family history of breast cancer, other known risk factors may differ in their contribution to overall breast cancer risk. The group of women with a family history of breast cancer, usually defined in epidemiological studies as one or more affected first-degree relatives, is genetically heterogeneous (see discussion on hereditary breast cancer genes below), which may have contributed to differences in outcome between these studies.

Colditz *et al.*[61] prospectively studied the impact of risk factors for breast cancer in 89,132 women with or without a history of breast cancer (approximately 6 % and 64 % respectively), who had been included in the Nurses' Health Study. The authors observed little or no protection from later age at menarche, high parity or early age at first birth in women with a family history of breast cancer, whereas these factors where associated with decrease in breast cancer risk in women without a family history.

In contrast, in the combined seven case-control studies (totalling 3152 cases and 4404 controls) analyzed by Andrieu *et al.*[62] and in the case-control study (6705 cases and 9341 controls) by Egan *et al.*[63] no clear differences in the effects of reproductive factors on breast cancer risk between women with or without a family history were observed, with the exception of high parity, which was associated with a stronger decrease in breast cancer risk in women with a positive family history in the Egan study. Katsouyanni *et al.* [64] found the collective impact of adult-life risk factors on breast cancer risk in women with a family history of breast cancer to be higher (RR 2.3) than in women without such a family history (RR 1.5).

Past use of oral contraceptives, use of postmenopausal hormones and history of benign breast disease showed similar relative risks between women with and without a family history of breast cancer in the Golditz study[61]. There is some evidence to suggest that use of oral contraceptives increases the risk (RR 7.8) of breast cancer in women carrying germline mutations in the breast cancer genes BRCA1 or BRCA2 (see below) more than in other women[65].

With respect to benign breast disease, Dupont et al.[66] observed that complex fibroadenomas increased breast cancer risk (RR 3.7) in women with a family history of breast cancer compared with relatives without such a fibroadenoma. In addition, Skolnick *et al.*[67] analyzed families with a clustering of breast cancer and suggested that genetic susceptibility may cause both proliferative benign breast disease and breast cancer in those kindreds.

Smoking, a possible breast cancer risk factor, was shown to be associated with a decrease in breast cancer risk (RR 0.46) in BRCA1 or BRCA2 mutation carriers (see below)[68]. Whether the carcinogenic effects of smoking are outweighed by its possibly (anti-oestrogenic ?) protective effects in these women, remains to be determined.

Hereditary breast cancer

Approximately 5-10 % of breast cancer patients in the general population are estimated to be carriers of a highly penetrant breast cancer susceptibility gene[3,4]. Hereditary disorders featuring breast cancer are summarized in the table below.

Table 3 Hereditary disorders associated with increased breast cancer risk

Name:	References:
Hereditary Breast (Ovarian) Cancer	(see main text)
Cowden syndrome	715,716
Peutz-Jeghers syndrome	195,717
Li-Fraumeni syndrome	402,403
Androgen-receptor gene germline mutations (male breast cancer)	718,719
Ataxia Telangiectasia (including heterozygosity)	16,106
Bloom syndrome	201
Fanconi Anaemia	202
Hereditary Non-Polyposis Colorectal Cancer (HNPCC)[1]	(see below)

1 = breast cancer has been reported in HNPCC as well as in its variant Muir-Torre syndrome. Whether there is a true association between HNPCC and breast cancer is controversial. Increased breast cancer risk was not observed in epidemiological studies on HNPCC[22,248,332,334] (with one exception[721]) and there is as yet no strong evidence for a significant increase in breast cancer risk in HNPCC families. Molecular study of a case of breast cancer occurring in a HNPCC family suggested it was indeed causally linked to the familial HNPCC gene mutation[720].

Of great importance has been the cloning of two genes responsible for the majority of hereditary breast (and ovarian) cancer: BRCA1[69] estimated to be mutated in approximately 45 % of hereditary breast cancer families[70] and BRCA2[71,72] mutated in an similar percentage. These percentages may, however, be overestimated, as they were calculated in large highly penetrant families[73-75]. Approximately 80 % of hereditary breast-ovarian cancer families can be attributed to BRCA1 mutations [70,76].

The chance of finding a BRCA1 or BRCA2 mutation in any given breast cancer patient ranges from less than 1 % to almost 90 %, depending on various patient and family characteristics[75,79,80], including the age of onset of her tumour[81-86], family history of cancer (*e.g.* number of relatives and with (male) breast cancer and ovarian cancer)[85-87], and ethnic (*e.g.* Ashkenazi Jewish origin) or geographical background, which may be associated with specific types of mutations [78,88-98].

In families with male breast cancer far more BRCA2 than BRCA1 mutations are found[73,77,78]. Approximately 4-20 % of male breast cancer cases are due to BRCA2 mutations and these cases are not necessarily associated with a family history of breast cancer[77,99-101].

Heterozygosity for ATM gene mutations (population frequency 0.2-1 %), possibly contributes significantly to the total female breast cancer burden. However, as yet, data are inconclusive with respect to the exact risks and overall contribution [16,17,102-112]. There is evidence for the existence of other breast cancer genes[113-117].

Cumulative risks of breast cancer in women with a BRCA1 mutation were initially estimated to be 59 % at age 50 and 82 % at age 70 in the highly penetrant families used for linkage analysis[70]. It has become clear that BRCA1 and 2 related breast cancer risk may be significantly lower in families with a lower number of affected women. The estimates for breast cancer risk at age 70 now range from 56 % to 87 % (95 % confidence interval ranges from 40 to 95 %) for BRCA1[20,23,118-120] and from 37 to 84 % (95 % confidence interval ranges from 22 % to 95 %) for BRCA2[120-122].

Individual cancer risk may depend on the specific type of mutation within BRCA1 or BRCA2, but it is difficult to predict the risk in individual cases[78,97,118,123-126]. Breast cancer risk for males carrying a BRCA2 mutation is approximately 6% (95 confidence interval 1.4%-25.6%)[127], for those carrying a BRCA1 mutation this risk is probably 1 % or less.

Histology of familial and hereditary breast cancer

If specific histological types of breast cancer would be typically associated with hereditary breast cancer, then they could serve as a clinical marker to identify this disorder. Although several studies have addressed this issue and *e.g.* medullary type breast cancer appears to be over-represented in BRCA1 carriers, it is as yet impossible to identify BRCA1 or 2 mutation carriers by the type of breast cancer they have developed[128-135]. Among women aged 30-49 years, a family history of breast cancer was associated with an increased frequency of ductal carcinoma-in-situ, and among those aged 50 and older it was associated with an increased frequency of both ductal carcinoma-in-situ and invasive breast cancer[136]. However, the presence of germline BRCA1 and -2 mutations was not investigated in this group and carcinoma-in-situ of the breast is probably not a good clinical marker to identify gene carriers in families with a BRCA1 or BRCA2 mutation[128,137,138].

Survival in hereditary breast cancer

The natural history of ductal carcinoma-in-situ and breast cancer in women with hereditary breast cancer is still largely unknown. Compared with sporadic breast cancer, increased, similar and decreased survival has been reported in familial and in BRCA1 or 2-specific breast cancer[139-149]. Most studies could not demonstrate large differences in survival between patients with BRCA1 or 2 associated breast cancer or sporadic cases. Hereditary breast cancer is generally associated with an increased risk of multiple primary tumours. A family history of breast cancer might therefore be expected to be associated with increased rates of local recurrence in conservatively treated breasts compared with sporadic cases. However, similar recurrence rates were observed in familial and non-familial cases[150-152] and, more importantly, the same applied to BRCA1 associated breast cancer[144]. Still, additional studies need to be performed before one can decide whether breast-conserving treatment (as opposed to radical mastectomy) is truly indicated in hereditary breast-ovarian cancer.

Surveillance in hereditary breast cancer

The value of screening for breast cancer in hereditary breast cancer families by mammography and physical (self) examination is unknown. On average, 50 % of the women with a BRCA1 mutation will develop this tumour before the age of 50[20] The higher frequency of dense breasts in the younger age group may hinder radiological interpretation and may therefore lower the sensitivity of mammography. Given the much higher risks of breast cancer in women from hereditary breast cancer families and possible differences in tumour biology as compared with the general population, we cannot simply extrapolate findings from general population screening to this high risk group.

Another issue to consider is the radiation risk of periodic mammography[154]. Although benefits may by far exceed radiation risk of periodic mammography in the general population, the opposite might possibly be true for hereditary breast cancer. Especially in women with germline mutations in ATM gene periodic screening by X-rays may carry increased risks of tumour induction[16,155]. Alternative methods for imaging the breast, for example magnetic resonance imaging, may therefore turn out to be a better option[156,157].

Few data are available on the actual outcome of periodic screening in hereditary breast cancer and, although some preliminary results suggest a benefit from screening, no firm conclusions at all can be drawn at this point [158-170]. Although enhanced imaging techniques are likely to increase tumour detection rates in hereditary breast cancer, it remains to be seen whether screening will actually lead to reduction of mortality and morbidity in these patients[171]. In general, guidelines for follow-up care of women with BRCA1 and BRCA2 mutations presently include monthly breast self-examination, annually or semi-annually breast examination by a clinician, starting at age 25-35, and annual mammography starting at age 25-35[172-174] (policy on screening for ovarian, colon and prostate cancer is discussed in sections 2.3.9., 2.3.4. and 2.3.11. respectively).

Prophylactic mastectomy

A similar lack of data exists with respect to prophylactic (contralateral) mastectomy[175-177]. Schrag *et al.*[178] used available data and estimates on hereditary breast cancer risk and effects of preventive options and calculated that,on average, 30-year-old women carrying a BRCA1 or BRCA2 mutation, gain 2.9-5.3 years of life expectancy from prophylactic mastectomy. However, gain in life expectancy is not necessarily the same as gain in quality-adjusted life years and that fact should be considered in the decision analysis too[179,180]. Hartmann *et al.*[181] retrospectively

studied a group of 639 women with a family history of breast cancer (including 214 high-risk women) who underwent bilateral prophylactic mastectomy. They demonstrated a reduction in incidence of breast cancer of at least 90 %. Only long-term follow-up after prophylactic surgery of a large cohort of women, with a genetic status defined by DNA analysis, will allow us to calculate the remaining breast cancer risks in the various categories of patients.

Chemoprevention

Chemoprevention of breast cancer is experimental. Ongoing trials using the anti-oestrogen tamoxifen and the retinoid fenretinide include women with a family history of breast cancer. The National Surgical Adjuvant Breast and Bowel project P-1 Study in the USA demonstrated a reduction of breast cancer risk by the use of tamoxifen[182]. However, most women included in this study did not have a family history suggestive of hereditary breast-ovarian cancer and their status with respect to germline BRCA1 or 2 mutations has yet to be determined. Tamoxifen is unlikely to prevent the occurrence of oestrogen-receptor negative (ER-negative) tumours[182]. This might mean that its protective effect in hereditary breast cancer families is limited, as breast cancers in these families appear to be often ER-negative[183-186].

2.3.3 Cervix Uteri

Relative risks

Familial clustering of cervical cancer has been reported[187-190]. An estimated 15 % of patients with this tumour have at least one affected first-degree relative[187]. Most details on familial risks were reported by Furgyik *et al.*[187], who estimated that having a first-degree relative with cervical cancer is associated with an RR of 3 to develop squamous cell cancer of the cervix, an RR of 10 of developing adenosquamous cancer at this anatomical site and no increased risk to develop cervical adenocarcinoma . No other studies with such detailed estimates have been published so far, so this study remains as yet unconfirmed. In the Swedish Twin study, monozygotic twins of women with cervical cancer in situ were shown to have an RR of 4.8 to develop the same condition, whereas the RR for dizygotic twins was 2.4[191]. An analysis of large Mormon families did not demonstrate increased risks of cervical cancer in women with an affected sister. However, an excess of cervical cancer was observed in first-degree relatives of patients with

early-onset bladder cancer (RR 2.32)[192]. A family history of cancer in general was no risk factor for cervical cancer in a Yugoslavian study[193]. A study of Finnish patients with borderline ovarian tumours and their relatives demonstrated an RR of 7.8 for cervical cancer in the mothers of index cases[194].

Cervical cancer in hereditary disorders

In women with Peutz-Jeghers syndrome, physicians should be alert to the occurrence of an aggressive type of cervical cancer: adenoma malignum[195-199]. Cervical cancer probably also belongs to the tumour spectrum of a small group of hereditary disorders: ectodermal dysplasia, congenital dyskeratosis, Bloom syndrome and Fanconi anaemia, which are associated with other types of mucosal cancer as well[200-203]. However, risk figures have not been reported. It is also unknown whether in any of these disorders, HPV infection is an important risk factor for cervical cancer as it is in the general population.

Surveillance

No data are available on the value of screening for cervical cancer in the setting of familial risk factors. Regular cervical smears may not detect all cases of adenoma malignum in Peutz-Jeghers syndrome, given the possible minimal atypia of these tumour cells[198,199,204]. Since there is also an increased risk for ovarian tumours in women with Peutz-Jeghers syndrome, cervical smears every 1 to 3 years, yearly endocervical curettage as well as yearly transvaginal ultrasound, in addition to careful physical examination, have been recommended in this syndrome[198,205]. Some authors have recommended the use of periodic colposcopy [199] and CT scans in these patients[204]. Presence of the other mentioned syndromes is at least an argument to participate in population screening programs and early cervical screening has been recommended for women with Bloom syndrome[200] and Fanconi anaemia [202]. Possibly, an increased sensitivity to HPV infections plays a role in familial cervical cancer[206-212], and screening for HPV infection might therefore be a valuable addition to normal cytological screening in familial cases. This remains to be investigated.

2.3.4 Colon and rectum

Relative risks and life-time risks

Approximately 15-25 % of patients with colorectal cancer have one or more affected relatives[213,214]. There is a large body of evidence to show that the risk of developing colorectal cancer is influenced by the number and degree of affected relatives, the age at which their tumours were diagnosed as well as by the tumour site [213-229]. In addition, risk of colorectal cancer appears to be increased by the presence of colorectal adenomas (the colorectal cancer precursor lesions) in relatives and *vice versa*[230-236]. Differences in risk between gender and type of first-degree relatives (sibs versus parents) have also been observed. Known dietary risk factors for sporadic colorectal cancer appear to influence colorectal cancer risk in individuals with a positive family history as well[215,237], and there is evidence to suggest that the family history of colorectal cancer increases colorectal cancer mainly in individuals with a high beef and ethanol intake[238].

Relative risks for CRC in individuals having 1 first-degree relative with colorectal cancer found are generally estimated between 2 and 3. RR's associated with having 1 first-degree relative with colorectal cancer diagnosed before the age of 45 are approximately 2 to 5. Additional affected relatives are associated with a further increase in relative risk. In terms of absolute risks, depending on the number and degree of affected relatives, cumulative life-time risks of 6 % (one 1st degree relative), 8 % (one 1st + one 2nd degree relative), 10 % (one 1st degree relative, diagnosed < 45 yr), 17 % (two 1st degree relatives) have been estimated[213,239]. Increased risk of colorectal cancer has also been observed for first-degree relatives of patients with leukaemia (RR 1.7-2.0, subtype not specified), cancer of the prostate (RR 1.3-1.48), breast (RR 1.3-1.7), cervix (RR 1.5), ovary (RR 1.3-2.7) and uterus (RR 1.4-1.6)[49,240-242].

Hereditary colorectal cancer

Colorectal tumours are a feature of a number of hereditary disorders which can be divided in those typically featuring many polyps (dozens, hundreds and even thousands) and those that do not [243-246](table 4). Without medical intervention, the life-time risk of colorectal cancer in some of these disorders is very high: up to 90 % in hereditary non-polyposis colorectal cancer (HNPCC) and virtually 100 % in familial adenomatous polyposis (FAP).

Hereditary non-polyposis colorectal cancer (HNPCC)

HNPCC is the most frequent hereditary disorder underlying colorectal cancer. It causes approximately 5 % of colorectal cancer cases. Also, it may be one of the more difficult common human cancer syndromes to recognize in the clinic. Therefore it deserves a more detailed discussion here. HNPCC is an autosomal dominant disorder characterized by a 70%-90 % cumulative life-time risk of - colorectal cancer, often with an early onset (average 40-50 years), proximal location (70 %), and an increased risk of extra-colonic tumours: particularly endometrial cancer, with a cumulative life-time risk of 30-50 %, ovarian cancer, stomach cancer, cancer of the small intestine and also cancer of the urinary tract [247-254] (see also table 1). Multiple primary colorectal tumours are a characteristic feature of this disorder.

Molecular aspects of HNPCC

At the molecular level, HNPCC is characterized by germline mutations in genes responsible for the repair of DNA replication errors, the mismatch repair genes. Several of these genes have been identified in the human and germline mutations in HNPCC patients have been found in 5 of them: hMLH1, hMSH2, hPMS1, hPMS2 and hMSH6[255-260]. Germline mutations in these genes have been detected in approximately half of the HNPCC families[261,262]. Deficiency of the first four of these genes causes replication errors (mismatches) in repetitive DNA segments, known as microsatellites. If the resulting microsatellite instability damages genes critical for cell growth control and other genes which contribute to genomic stability, this in turn may lead to tumour development[263-266]. Loss of other functions of the mismatch repair genes, especially those involved in the repair of DNA damage caused by environmental agents, might be important for tumorigenesis as well [264,267-269]

It has been shown that microsatellite instability is not a unique feature of tumours in HNPCC patients[270], but occurs in different percentages in the majority of sporadic solid neoplasms[271]. Nevertheless, finding the presence or absence of microsatellite deficiency, especially in colorectal tumours, may help in deciding for clinical purposes whether to screen for germline mutations in certain MMR genes or not[272].

Table 4 Hereditary disorders associated with colorectal tumours

Disorders with polyposis :	**References**
Cowden syndrome (H/A/Hyp, sometimes G)[1]	663,722,723
Familial Adenomatous Polyposis (A)[2]	10,724
(Familial Giant Hyperplastic Polyposis)(A/Hyp)	674
(Hereditary Mixed Polyposis syndrome)(H/A/Hyp)	681,725,726
Juvenile Polyposis (H, sometimes + A)	317,498
Peutz-Jeghers syndrome (H, sometimes + A)	195,204,205
(Turcot syndrome)(A)[3]	706,707,727
Disorders without polyposis:	
Attenuated Familial Adenomatous Polyposis (A)[2]	724,728
Bloom syndrome (A)	201,657
Congenital Dyskeratosis (A)	586
Familial Gastrointestinal Stromal Tumours (ST)	673
Hereditary Breast(Ovarian) Cancer (BRCA1/BRCA2) (A)[4]	23,97
Hereditary Non-Polyposis Colorectal Cancer (A)[5]	252,253
Li-Fraumeni syndrome/germline TP53 mutations (A)	402,729
(Punctate Palmoplantar Keratoderma) (A)	701,702
X-linked Agammaglobulinaemia (A)	712
X-linked Lymphoproliferative disease (NHL)	18
Blue Rubber Bleb Nevus syndrome (Haem)	599
Carney Complex (Schw)	659
Gorlin syndrome (Basal Cell Nevus syndrome)(H)	8,730
Multiple Endocrine Neoplasia, type 2B (G)	693,731
Neurofibromatosis, type I (N/G)	698,732
Tuberous Sclerosis (H/A)	733,734

Legends table 4

Polyposis = these disorders typically present with dozens to hundreds of polyps;

Syndrome name between ( ) = status as separate inherited disorder as yet unclear;

Conditions in italics: risk of malignant colorectal tumours (probably) not increased;

Between ( ), after syndrome name: type of associated benign colorectal lesions: A = adenoma; G = ganglioneuroma; H = hamartoma; Haem = haemangioma; Hyp = hyperplastic polyp; N = neurofibroma; NHL = intestinal Non-Hodgkin lymphoma; Schw = schwannoma; ST = gastrointestinal stromal tumour.

1 = Includes Bannayan-Riley-Ruvalcaba syndrome [651,735,736].

2 = familial adenomatous polyposis (FAP) includes Gardner syndrome (name used to refer to FAP with manifestations outside the gastrointestinal tract). In a subgroup of FAP families with specific types of germline mutations in the APC gene, the number of polyps is low, or at least appears to be low if the colonic mucosa is not dye-sprayed during endoscopy. Polyps and carcinomas are also located more proximal in the colon as compared with regular FAP and often present at a later age. This variant is termed attenuated familial adenomatous polyposis (AFAP). A familial hypermutatable tract in the APC gene has also been implicated in familial clustering of (non-polyposis) colorectal cancer, but has in comparison only a minor effect on cancer risk[737-739].

3 = some cases have either FAP or HNPCC.

4 = increased colorectal cancer risk is not associated with the BRCA1 and 2 mutations common among Ashkenazi Jews[740].

5 = Includes Muir-Torre syndrome[593,741]

HNPCC criteria

The classical criteria of HNPCC, known as the Amsterdam criteria, were initially developed for research purposes and did not include these molecular data. These criteria demanded: (a) the presence of at least 3 relatives with histologically verified colorectal cancer, one of the relatives being a first-degree relative of the other two, (b) colorectal cancer involving at least 2 successive generations, (c) at least one case of colorectal cancer diagnosed before age 50 and (d) exclusion (on clinical grounds) of Familial Adenomatous Polyposis.

These criteria have been criticized as being to strict for clinical purposes and justly so, because they did not take into account the wide range of extra-colonic tumours associated with HNPCC and presence or absence of tumour microsatellite instability[273-276]. Germline mutations in the HNPCC genes have indeed been demonstrated in patients with early onset colorectal cancer, featuring microsatellite instability in their tumours, in the absence of a strong family history of colorectal cancer[277]. Although cancer risks associated with these mutations found outside classical HNPCC families may differ from those found in families fulfilling the Amsterdam HNPCC criteria, these risks may still be high, especially if those families were ascertained through young colorectal cancer patients[254]. It might therefore be useful to look for microsatellite instability in colorectal tumours of patients that do not meet the Amsterdam criteria.

Guidelines for selection of tumours to be tested, referred to as the Bethesda guidelines, have recently been published[278] (table 5). Once microsatellite instability is found in colorectal tumours, then testing the patients with those tumours for germline HNPCC gene (mismatch repair genes, MMR) mutations is indicated. If microsatellite instability is absent in colorectal tumours (also in those of other relatives in that family) then this does not exclude HNPCC[279]. It appears that germline HMSH6 mutations are associated with the absence of microsatellite instability and testing for this gene should therefore be considered in patients with possible HNPCC and absence of microsatellite instability in colorectal tumours.

Table 5 Bethesda selection guidelines for testing of tumour microsatellite instability (modified after Rodriguez-Bigas *et al.* [278])

1. individuals with cancer of any type at any age, in families meeting the Amsterdam criteria.

   Comment: preferably, colorectal tumours should be tested, or, if not available, any HNPCC associated extra-colonic tumour (endometrial, ovarian, gastric, hepatobiliary, small-bowel cancer, transitional cell carcinoma of renal pelvis or ureter).

2. individuals with colorectal or endometrial cancer diagnosed at age <45

   Comment: any colorectal cancer type, but especially tumours with > 50% signet ring-cell-type, or right-sided location and undifferentiated histological pattern

3. individuals with colorectal adenomas diagnosed at age <40

4. individuals with two HNPCC related cancers diagnosed at any age: colorectal cancer or any of the HNPCC associated extra-colonic tumours (listed under item 1.)
   Comment: preferably the colorectal tumour should be tested

5. individual with colorectal cancer at any age + a first-degree relative with colorectal cancer and/or colorectal adenoma and/or HNPCC related extra-colonic cancer (see 1. for listing); one of these cancers diagnosed at age <45 or adenoma at age <40

   Comment: preferably the colorectal tumour should be tested.

Purpose of the Bethesda guidelines is to select individuals for screening for germline mismatch repair gene (MMR) mutations by first looking at presence or absence of tumour microsatellite instability (MSI)(methods reported by Bolland et al.[742]). From the mentioned selection criteria, the presence of right-sided mucinous colorectal cancer diagnosed before the age of 45 appears to be a particularly useful predictor of microsatellite instability[743]. Individuals/families with MSI-high tumours should subsequently be tested for germline hMSH2 and hMLH1 mutations (hPMS1 and 2 mutations are probably very rare). In the selected individuals/families which appear to have MSI-low tumours, testing for germline hMSH6 gene mutations should be considered.

Survival in HNPCC

Although colorectal cancer in HNPCC at the histological level is often of a mucinous, highly proliferative type[252], which is generally associated with less favourable prognosis, survival in HNPCC appears to be better than in sporadic colorectal cancer cases with similar staging[280-283]. However, studies by Percepe *et. al.* and Bertario *et al.* showed no difference in survival[284,285]. These opposite findings might possibly be explained by heterogeneity in the inherited genetic background and in the somatic mutation spectrum of the tumours.

Surveillance in familial and hereditary colorectal cancer

A range of screening schedules in familial colorectal cancer has been developed, as shown in table 6. It is presently unclear if, and how, these screening protocols for colorectal cancer should be modified by the presence or absence of microsatellite instability in colorectal tumours of for example single affected relatives, where no HNPCC mutation can (yet) be identified[286-289]. In patients with a high risk of developing colorectal cancer, the use of endoscopy rather than radiological examination or biochemical testing of the faeces[290] has been favoured since it allows for direct inspection of the anatomical site at risk and simultaneously sampling or removal of the polyps. Colonoscopy is preferred to sigmoidoscopy given the frequent presence of proximal tumours in patients with an increased risk, which may occur in the absence of distal tumours[231,291-293].

The possibility to detect and remove colorectal tumours in their pre-cancerous state makes screening for them potentially attractive. However, whether morbidity and mortality are significantly reduced by screening in individuals at increased genetic risk of colorectal cancer is as yet largely unknown. In FAP though, the case for screening (and prophylactic surgery) has been made[294]. In HNPCC there is also evidence that screening, which has to be meticulous[295], indeed reduces colorectal cancer rates[296-298]. No studies have yet demonstrated the assumed benefits of screening in Peutz-Jeghers syndrome and juvenile polyposis.

Chemoprevention

Chemoprevention and dietary intervention in familial and hereditary colorectal cancer is experimental[299,300]. Use of aspirin has been recommended in familial colorectal cancer[301] given its effects on the occurrence of sporadic cases[302] but its effect has not yet been established in individuals with a family history of colorectal cancer. The effects of vitamin C plus vitamin E with or without a high dose wheat bran fibre supplement on polyp development in FAP has been studied in a double-blinded, placebo controlled trial, as has been the effect of sulindac[303,304]. Sulindac and wheat bran fibre were each shown to reduce polyp growth. The protective effects of aspirin and resistant starch in FAP and HNPCC are currently being studied in double-blind trials, known as the CAPP 1[305] and CAPP 2 study[306], respectively. Calcium has been studied for its protective effect in both HNPCC and FAP. Results in both conditions were not promising[307,308]. Sulindac has been shown to induce regression of colorectal adenomas in FAP[304,309]. It has been used occasionally in Juvenile Polyposis where its role as a chemoprotective agent is as yet unclear[310].

Prophylactic surgery and gene therapy

Prophylactic colectomy is the general procedure in FAP, with most patients being in their twenties at the time of surgery[10,311-313] Performing prophylactic colectomy in HNPCC is controversial[314-316]. This is to be expected given the fact that within HNPCC heterogeneity of risks of colorectal cancer and extra-colonic tumour exists and the exact impact of screening and prophylactic surgery for colorectal cancer in this disorder still needs to be determined. Prophylactic colectomy has also been proposed for patients with juvenile polyposis[317]. For the distant future, gene therapy of the gut may be an alternative approach to tumour prevention[318]. Liposomal delivery of wild type APC genes to the colonic epithelium has been studied in a mouse and rat FAP model. Although the level of gene uptake was high, levels of APC expression were low and did not persist beyond three days[319].

Table 6 Screening guidelines for colorectal tumours in asymptomatic individuals with a family history of colorectal cancer and adenomas

family history	method	screening start (yr)	end (yr)	interval (yr)	remarks	references
●1-2 x FDR and/or	FOBT	40		1	Expert Panel[1]	[744]
1 FDR adenomatous	sigmoidoscopy	40		5	-	
polyp < 60	BE	40		5-10		
	colonoscopy	40		10		
●1 x FDR< 45 or	colonoscopy	25		5	St Mark's	[745, 239, 633]
2 x FDR						
●1 x FDR> 45	FOBT	25		1	-	
●1 x FDR	FOBT	35-40		1	WHO;	[746]
	DRE, sigmoidoscopy	35-40		3-5	minimal options	
	or colonoscopy					
●1 x FDR<55 and/or	FOBT	35-40		1	-	
≥ 2 x FDR	colonoscopy	35-40		3-5		
●HNPCC*	colonoscopy	20-25		2	ICG-HNPCC	[335]
	colonoscopy	20-30	40	1-2	Expert Panel[1]	[744,747]
		40		1	-	
	colonoscopy	20-25		1-3	Cancer Genetics Studies Consortium	[748]

•FAP*	(BE or) sigmoidoscopy,	10-15	30	1-2		200
	(or colonoscopy)	30	40**	2-3		
•Attenuated FAP	colonoscopy	20		1		682
•Peutz-Jeghers*	colonoscopy (or	10-20		1-2		205,682,749
	sigmoidoscopy +			3		749
	FOBT)			1		
•Juvenile Polyposis*	colonoscopy (or	10-20	40**	3-5		498,682
	sigmoidoscopy +			3		749
	FOBT)			1		
•BRCA1 mutation carriers*	follow guidelines for the general population				Cancer Genetics Studies Consortium	172

Unless stated otherwise, family history refers to relatives with colorectal cancer, stating degree of kinship and age at diagnosis of tumour. Once benign or malignant tumours are detected by screening, guidelines for follow-up after treatment usually differ from those presented in this table, depending on the diagnosis and type of treatment. The reader is referred to the references for further details. With respect to the human cancer syndromes, the screening schedules apply to proven gene carriers as well as to relatives with a 50 % risk of carrying the gene (e.g. from families where the mutation has not yet been found).

BE = barium enema; DRE = digital rectal examination; FDR = first-degree relative (parents, siblings, children); FOBT = faecal occult-blood testing; SDR = second-degree relatives (grandparents, siblings of parents, children of siblings and grandchildren).

** = In these hereditary disorders screening for extra-colonic tumours is also performed (see references for details).*

*** = in patients/gene carriers, screening will continue.*

[1]) = the clinical practise recommendations of this panel are endorsed by a number of organisations including the American Cancer Society, American College of Gastroenterology and American Gastroenterological Association.

2.3.5 Endometrium

Relative risks

The proportion of endometrial carcinoma based on a strong genetic predisposition has been estimated at less than 1% to 6 %[320-322]. The risk of endometrial cancer has been found to be positively associated with a family history of this cancer[323]. Women with a first-degree relative with endometrial cancer were shown to have an RR of 1.5-2.8 of developing this tumour type[241,320,322,324], although in a study of data from the Utah Population Database, first-degree relatives of women with endometrial cancer failed to show a significant excess of this tumour type[49]. This difference may be due to differences in genetic background and exposure to risk factors between the Mormon population monitored in the Utah database and other populations. An increased risk of endometrial cancer was found in first-degree relatives of patients with colorectal cancer (RR 1.8)[322] and ovarian cancer (RR 1.1)[240] and excess mortality from endometrial cancer was observed in first-degree relatives of breast cancer patients (standardized mortality ratio 166 %)[325].

Hereditary endometrial cancer

Endometrial cancer is a frequent feature of HNPCC (see section 2.3.4.) and a strong indicator for this syndrome in the presence of colorectal cancer[262]. Although some authors have suggested the existence of hereditary site-specific endometrial cancer based on striking cases of familial clustering of this tumour[326-328], these cases are possibly variants of HNPCC[329,330]. Different figures have been published on the cumulative life-time risk of endometrial cancer in HNPCC, ranging from 20% by age 70[331] to 39 % by age 70 and 43% by age 80[332]. A more recent study analyzed HNPCC families for germline mutations in hMLH1 and hMLH2, and calculated cancer risks only in proven mutation carriers. Life-time risks of endometrial cancer of 42 %-61 % were found in the carriers of hMLH1 and hMSH2 mutations[251]. Endometrial cancer in HNPCC may be diagnosed as early as in the late-twenties; the mean age at diagnosis was calculated at 48 years (range 27-72 yr) in a study by the International Collaborative Group on HNPCC[333]. Endometrial cancer appears not to be equally prevalent in all HNPCC families[334]. Causes of this unequal distribution remain, however, to be identified.

Preventive options

In HNPCC, screening for endometrial cancer by physical examination and transvaginal ultrasonography is recommended, starting at age 30-35 with 1-2 years interval[335]. As yet, the effect of this type of screening on mortality and morbidity in these families is unknown. In families with a clustering of endometrial cancer not fitting the clinical HNPCC criteria, screening for endometrial cancer following the above-mentioned schedule might be considered[329]. Prophylactic hysterectomy is considered in HNPCC patients, with completed family, already undergoing surgery for colorectal cancer[252].

2.3.6 Kidney

Relative risks

Having one first-degree relative with kidney cancer (type unspecified) was observed to be associated with an RR of 1.6 of renal cell cancer in the International Renal-Cell Cancer Study, in which 1774 renal cell cancer patients and 2359 controls participated[336].

Hereditary renal cancer

In less than 1-2.5 % of cases, renal cell cancer occurs in the setting of a hereditary cancer syndrome[5,6]. In von Hippel-Lindau disease (VHL), (non-papillary) clear cell carcinomas of the kidney occur in approximately 26 % of the patients, with a mean age at presentation of 35-40 years[337]. This accounts for about 50 % of deaths in VHL[337]. Renal clear cell carcinomas with a striking clustering in families without the additional features of VHL have also been reported[338,339]. Constitutional chromosomal translocations between chromosome 3 and either 2, 6, 8 or 12 have been observed in some of these families and individual patients with renal cell cancer[340-344]. The 3;8 translocation was found to have caused a fusion between the FHIT and TRC8 genes[345].

It has been postulated that the clustering of cases in the families without translocations is caused by an autosomal dominant disorder, different from VHL, referred to as familial renal cell carcinoma or familial non-VHL non-papillary clear cell renal cancer [338,346]. As the name implies, renal cell carcinomas of the papillary type are the feature of hereditary papillary renal cell carcinoma[347-350], associated

with germline MET gene mutations. In tuberous sclerosis (TS) angiomyolipomas of the kidney are found in approximately 49 % of the patients[351].

Renal cell carcinoma is found in approximately 2 % of the TS patients[351] and these tumours have been reported as either oncocytomas, clear cell or anaplastic types[352,353]. Transitional cell carcinomas of the renal pyelum and ureter may occur in hereditary non-polyposis colorectal cancer (HNPCC; see also sections 2.3.4 and 2.3.5)[251,332,334,354]. In hereditary hyperparathyroidism-jaw tumour syndrome[355] and in Perlman syndrome renal hamartomas have been documented[356].

Wilms tumour in hereditary disorders

Wilms tumour (nephroblastoma) is the most common renal tumour in children. Approximately 0.5-2.5 % of cases is familial[357-360]. The proportion of bilateral cases is higher in familial than in sporadic cases (20 % versus 3 %)[358]. No clear evidence for cancer prone families, *i.e.* families with an increased risk to develop cancer in general, was found in the French Wilms tumour study, which looked at relatives of 501 patients[360]. However, site-specific familial Wilms tumour has been reported, although the genetics of this presumably autosomal dominant disorder are unclear[361-368]. Wilms tumour also occurs in association with Beckwith-Wiedemann syndrome[369], Perlman syndrome[369] and Simpson-Golabi-Behmel syndrome[370], which can all be classified as "overgrowth" syndromes[371,372]. The tumour has also been found in association with WAGR syndrome (Wilms tumour, Aniridia, Genitourinary anomalies, Retardation) and Denys-Drash syndrome[371].

Surveillance

Annual screening for renal cell cancer by urine testing, ultrasound and computer tomography is advised in VHL, from age 16-20[338,373]. Screening is also recommended in familial clustering of non-VHL renal cell cancer, following the VHL protocol[338]. The renal tumours in carriers of the Hereditary Papillary Renal Cancer gene (MET) can easily be missed by ultrasound screening and enhance poorly on CT. Screening these gene carriers with contrast-enhanced CT or MRI is therefore recommended[374]. In HNPCC families with a positive family history of urinary tract cancer, screening by ultrasound and urine analysis every 1-2 years is recommended, starting at age 30-35[335], although excretory urography might be a better option[375].

Screening for Wilms tumours by ultrasound and urinalysis at 3-month intervals until age 6-7 has been recommended in Beckwith-Wiedemann syndrome, WAGR, Perlman, Bloom syndrome and Simpson-Golabi-Behmel syndrome[371,376]. However, little is known of the effects of screening on renal cell cancer or Wilms tumour morbidity and mortality in the above mentioned disorders.

2.3.7 Lung

Relative risks

A positive family history of lung cancer has been documented in approximately 12-23 % of lung cancer patients[377-379]. A (smoking-adjusted) RR of 2.4 to develop lung cancer in general, and an RR of 4 to develop adenocarcinoma of the lung, has been estimated in first-degree relatives of lung cancer patients[379,380]. A relative risk of 5.31 for offspring of lung cancer patients has been reported[381]. Although differences appear to exist between risks associated with the various histological types of lung cancer, the studies on this subject are contradictive[378,382,383].

Although familial clustering of lung cancer might be explained by shared exposure to external risk factors, particularly tobacco smoke, there are data which suggest that refraining from (active and passive) smoking does not normalize lung cancer risk in case of a positive family history. An affected first-degree relative with lung cancer has been shown to be associated with an RR of 6.1. among non-smokers in the 40-59 year old age group, after adjusting for environmental smoke exposures[377]. An RR (1.5) to develop the lung cancer subtype adenocarcinoma was found among non-smoking first-degree female relatives of lung cancer patients[384]. However, another study did not show a significant increase of lung cancer risk among non-smokers with a family history of lung cancer[385].

Genetic (segregation) analysis of smoking-associated malignancies suggest that Mendelian factors may influence the risk of cancers that are known to be smoking associated[386]. In this model, approximately 15 % of the population could be expected to carry an allele of some gene with an increased susceptibility to smoking-associated cancer. These carriers were estimated to have a mean age-of-onset of approximately 22 years earlier than non-carriers[386]. In contrast, twin studies did not indicate the existence of a strong genetic lung cancer predisposition[191,387].

Segregation analysis of the Louisiana lung cancer dataset supported a high-risk allele frequency of 2% and carriers were estimated to have an RR of 17.3 to develop lung cancer[388]. A genetic epidemiologic study in the families of non-smoking lung cancer probands rejected all simple Mendelian models when trying to explain lung cancer aggregation in these families[389]. It should not come as a surprise that all these findings are difficult to interpret, given the fact that many genetic polymorphisms have been shown to be associated with differences in lung cancer risk which may have complex interactions with various environmental risk factors[390-398].

Not only familial lung cancer, also familial mesothelioma has been reported and is possibly caused by an inherited increased susceptibility, in this case to asbestos exposure[399-401].

Lung tumours in hereditary disorders

Lung cancer may occur as part of Li-Fraumeni syndrome[402,403] (which may even present with multiple synchronous lung cancers[404]), idiopathic pulmonary fibrosis (Hamman-Rich syndrome)[405] and fibrocystic pulmonary dysplasia[406]. Lymphangioleiomyomatosis of the lung is a feature of Tuberous Sclerosis[407,408]. Carcinoid tumours of the lung (and other organs) are part of the Multiple Endocrine Neoplasia type 1 phenotype[409]. Pulmonary hamartomas have been documented in Cowden syndrome[410].

Preventive options

It has been suggested that periodic chest radiographs may be appropriate for individuals at high risk of lung cancer[411], but their value remains to be proven for this population. Chemoprevention of lung cancer is experimental [412]. Refraining from smoking may be especially important in the presence of a family history of smoking-associated tumours, including lung cancer. However, as discussed above, familial risk factors independent of smoking may very well exist.

2.3.8 Nasopharynx/oropharynx/larynx

Relative risks

A family history of head and neck aero-digestive tract squamous cell carcinomas increases the risk to develop these tumours[413]. Relative risks for first-degree relatives of patients with these tumours have been estimated between 3.5 (affected

first-degree relatives in general) and 14.6 (siblings)[414-416]. The high risks for siblings, compared with first-degree relatives in general, may hint at the existence of alleles associated with high susceptibility if they are inherited from both parents. This would fit the outcome of a segregation analysis of the clustering of nasopharyngeal carcinoma in Taiwanese families, which suggested the existence of such alleles[417]. Tumour risk is especially elevated in relatives of patients with multiple primary head and neck tumours (RR 7.9)[415]. First-degree relatives of lung cancer patients are at an increased risk to develop nasal cavity/sinus, mid-ear and laryngeal cancer (RR 4.6)[418]. An excess mortality from cancers of the larynx has been documented in first-degree relatives of breast cancer patients (standardized mortality ratio 177 %)[325].

Head and neck cancer in hereditary disorders

Head and neck cancer has been found in 18 % of patients with Bloom syndrome[419]. It is also part of the phenotypes of Congenital Dyskeratosis[203] (including Keratitis Ichthyosis Deafness (KID) syndrome which is associated with tongue cancer[420]), Fanconi anaemia[202], recessive Dystrophic Epidermolysis Bullosa[421] and Xeroderma Pigmentosum (especially of the tongue)[422].

Preventive options

Especially in patients with Bloom syndrome, Congenital Dyskeratosis and Xeroderma Pigmentosum, where risks of head and neck cancer are relatively high, screening for these tumours might be warranted. Screening upper endoscopy in a larger group of individuals at risk has also been proposed[423]. However, data on the clinical impact of head and neck cancer screening in the mentioned hereditary disorders or general population are lacking. Chemoprevention, using retinoids, is still experimental, and has not yet been specifically aimed at individuals with high genetic risks[424,425].

2.3.9 Ovary

Relative and life-time risks

A family history of ovarian cancer has been documented in 1-7 % of patients with this tumour[426-428]. The estimations of the proportion of hereditary cases range between less than 1 % and up to 20 %[4,426,429-431], with most references favouring the range of 5-10 %. First-degree relatives of ovarian cancer patients have an RR of developing this tumour of approximately 3-5[240,428,432-436].

The risk to mothers (RR 1.1) is lower than those to sisters (RR 3.8) and daughters (RR 6.0) of affected women[436]. Second degree relatives have an RR of 2-3, and third degree relatives of affected patients have an RR of approximately 1.5 to develop ovarian cancer[430,434].

Among Jewish women, who have been reported to be more likely than non-Jewish women to carry mutations in the hereditary breast-ovarian cancer genes BRCA1 and 2, the relative risk for first-degree relatives of affected women is higher: 8.8[437]. A cumulative life-time risk of developing ovarian cancer of 3-6 % has been estimated for first-degree relatives and 4-5% for second degree relatives[426,436]. More specifically, the risk for daughters is approximately 7% and for sisters of an affected woman is approximately 5 %[436]. Additional affected relatives increase the risk: having two affected first-degree relatives is associated with a risk of approximately 20 %[438].

A decrease in age at diagnosis in the index case is associated with an increase in risk[428,432]. However, this trend has not been observed in all studies[429,439]. One study, which tried to correct for ascertainment bias due to self-referral, indicated a decrease in age at diagnosis from generation to generation (known as 'anticipation') in families with ovarian cancer[440]. Having a first-degree relative with cancer of any of the following organs is associated with an increased ovarian cancer risk: endometrium (RR 1.9)[430], pancreas (RR 2.9)[430] or breast (1.6-1.7)[324,430] colon and rectum (RR 1.1-3)[430,441], prostate (RR 5.8)[441], lung (RR 3.4)[441] or lip (RR 1.9)[49]. Granulosa cell tumours appear to be strongly associated with a family history of breast (RR 17.4) or uterine cancer (RR 16.8)[430]. Compared with breast cancer, bilaterality of ovarian cancer is probably not as strong an indicator of genetic predisposition[442].

Histology of familial and hereditary ovarian cancer

All histological types of ovarian tumours have been observed in familial clusters[443-446]. However in familial ovarian cancer, serous cystadenocarcinomas are over-represented and mucinous adenocarcinomas are under-represented compared with sporadic cases[429,443,447]. Some studies have indicated an association between a family history of breast, endometrial cancer or bone cancer and the occurrence of, in particular, endometrioid ovarian cancer[430,448]Whether a family history of borderline ovarian cancer is a risk factor for ovarian cancer is as yet unclear[429,443,449,450].

Hereditary ovarian cancer

Ovarian cancer may occur in a number of hereditary disorders. The most frequent of these is Hereditary Breast-Ovarian Cancer. This syndrome is often clinically defined as the presence of at least 3 cases of breast and/or ovarian cancer in at least two successive generations, with one affected relative being a first-degree relative of the other two (or a second degree relative in case of the apparent transmission of the gene through the paternal line).

In 80 % of (breast-)ovarian cancer families meeting these criteria, germline mutations in the BRCA1 gene are present[69,70,120]. The proportion of ovarian cancer due to germline BRCA1 mutations is estimated at 4.4-5 % in general[451-453] and 5.7 % below age 40 at diagnosis, 4.6 % between ages 40 and 49, and 2.1 % between ages 50 and 70 at diagnosis[87]. Women in Hereditary Breast-Ovarian Cancer families carrying a mutant BRCA1 gene were initially estimated to have average cumulative risks of developing ovarian cancer of below 1 % by age 40 and approximately 63% by age 70[20]. However, there is significant evidence of heterogeneity with regard to this risk and it may be considerably lower in women ascertained through families with relatively little cases of breast and ovarian cancer[123,452]. This complicates counselling. Estimates of cumulative ovarian cancer risk by age 70 now range from 16 % to 63 % (borders of the 95 % confidence intervals range from 6 % to 80 %) for BRCA1[20,23,118] and 27 %(95 % confidence interval ranges from 0 % to 47 %) for BRCA2[120]. In BRCA1 mutation carriers, the risk of ovarian cancer decreases with increasing age at last childbirth and, contrary to sporadic ovarian cancer, increases significantly with increasing parity[454]. As mentioned above, mucinous ovarian cancer is under-represented in familial ovarian cancer in general and the same appears to be true for BRCA1-associated ovarian cancer[455]. Borderline ovarian cancer also appears to be relatively under-represented in BRCA1 mutation carriers and is therefore a poor indicator of the presence of a germline BRCA1 or BRCA2 mutation[452,456-459].

In approximately 10-14 % of Hereditary Breast-Ovarian Cancer cases, BRCA2 gene mutations are present[120,460], which confer a risk of ovarian cancer of 0.4 % by age 50 and 27 % by age 70[120,460].

It remains to be seen whether a clinically important association between ovarian cancer risk and the type of BRCA1 or BRCA2 mutation exists[123-125].

Recent data show that, as compared with sporadic ovarian cancer cases, ovarian cancer associated with BRCA1 or 2 germline mutations may have a more favourable clinical course[461,462].

However, the opposite has also been reported [463-465]. Obviously, prospective studies in well defined cohorts are needed to settle this issue.

Ovarian tumours also occur in families with hereditary non-polyposis colorectal cancer[334], Peutz-Jeghers syndrome (mainly sex cord tumours with annular tubules (SCAT), and cystadenomas)[466], basal cell nevus (Gorlin) syndrome (mainly fibromas and fibrosarcomas)[7], ataxia telangiectasia (gonadoblastoma, dysgerminoma, fibroadenoma)[467], Frasier syndrome[468] and Denys Drash syndrome[469] (in both cases gonadoblastomas).

Surveillance in familial and hereditary ovarian cancer

Screening for ovarian cancer in women with an increased risk of developing ovarian cancer, based on family history and/or the proven presence of certain germline mutations, has been performed, usually starting at age 30-35, often by yearly transvaginal ultrasonography and serum CA-125 measurement, and sometimes combined with colour Doppler imaging [470-475]. Although the yield of such screening may be higher than in the general population, it is presently unclear whether these women actually benefit from it[476].

Prophylactic surgery

Prophylactic oophorectomy should be considered in high-risk women[426,477-479]. Given the low risks to develop ovarian cancer before the age of 35-40, this procedure is often performed after this age. Unfortunately, an increased risk of peritoneal carcinoma remains after surgery [480-484]. This risk has been estimated between 2 to 11 %, but the precise risks are unknown, in particular for the subgroups of patients with or without proven germline mutations in BRCA1[480,482].

Schrag *et al.*[178] calculated, using available estimates of ovarian cancer risk and effects of prophylactic surgery, that 30-year-old women who carry BRCA1 or BRCA2 mutations gain 0.17-1.7 years of life-expectancy from prophylactic oophorectomy, which could be delayed until age 40 with little loss of life expectancy. However, gain in life expectancy is not the same as gain in quality-adjusted life years and that fact should be considered in the decision analysis too[179,180]. With respect to chemoprevention, the use of hormonal contraceptives have been reported to lower ovarian cancer risk in the general population[485] as well as in a cohort of women with BRCA1 or BRCA2 germline mutations[486]. The latter findings need to be confirmed and the potential adverse effect of oral contraceptive use on breast cancer risk needs to be addressed[487].

2.3.10 Pancreas

Relative risks

There are a number of reports of familial clustering of pancreatic cancer[488-490]. In a series of 81 pancreatic cancer patients, 7 (9 %) had a family history of pancreatic cancer[491]. First-degree relatives of patients with pancreatic cancer were shown to have an RR ranging from 1 to 5.3 to develop this tumour[489,492,493]. Pancreatic cancer risk was also increased for mothers of daughters with borderline ovarian cancer (RR 4.9)[194] and for male first-degree relatives of breast cancer patients (RR 1.7)[494].

Pancreatic cancer in hereditary disorders

Germline mutations may play a significant role in perhaps 5 % of the total pancreatic cancer burden[488]. Possibly site-specific hereditary pancreatic cancer exists and in some of these families insulin dependent diabetes mellitus may act as a marker for pancreatic cancer[488,495-497]. Pancreatic adenocarcinomas may develop as part of a number of hereditary disorders[488,489,498,499]: hereditary non-polyposis colorectal cancer, juvenile polyposis, hereditary breast-ovarian cancer (BRCA2 type), familial atypical multiple mole melanoma syndrome, von Hippel-Lindau disease, Li-Fraumeni syndrome, hereditary pancreatitis and cystic fibrosis[499]. Germline BRCA2 mutations have been detected in pancreatic cancer patients without a family history suggestive of hereditary breast (ovarian) cancer (see section 2.3.2)[500,501]. In multiple endocrine neoplasia type 1 as well as in von Hippel-Lindau disease, endocrine pancreatic tumours occur[502,503].

Preventive options

In members of families with a striking clustering of pancreatic cancer (possibly hereditary pancreatic cancer) Lynch *et al.*[488] recommend the following diagnostic approach: yearly biochemical screening (alkaline phosphatase, amylase, lipase, carcinoembryonic antigen, CA-19-9) and abdominal CT scan. At the mean age of pancreatic cancer development in the high-risk family, endoscopic ultrasonography and endoscopic retrograde cholangiopancreatography for cytology and biomarker (K-ras) analysis are recommended. Ages at which to start or to end screening are not specified in their guideline. Prophylactic pancreatectomy has been performed in a few cases[490,497]. The value of screening and prophylactic surgery in high-risk families is as yet unknown.

2.3.11 Prostate

Relative risks

Prostate cancer is familial in approximately 15-20 % of cases[504,505]. A family history of prostate cancer is an established risk factor for this tumour[506-511]. Sons of an affected father have been shown to have RR's of developing this tumour ranging from 1.2 to 3.8, and RR's for men having an affected brother range from 1.9 to 4.7 [192,242,505,512-520]. Additional affected relatives further increase this relative risk, which may be as high as 11 in the presence of 4 affected relatives[512,521]. A relatively young age at diagnosis in first-degree relatives is another risk modifier: age 45-49 is associated with an RR of 3.18-16.6[242,520], 50-54 with an RR 2.6-3.5[242,520], 0-60 with RR 5.3[517], 0-65 with RR 5.9[192], and 0-70 with RR 2.3-3.4[242,522].

Monozygotic twins of prostate cancer patients have been shown in the Swedish twin study to have a 6-fold risk to develop prostate cancer compared with dizygotic twins[191,523]. These difference in concordance were confirmed by the American NAS-NRC Twin Registry[524] and support the notion of a hereditary prostate cancer predisposition.

According to some studies, having a first-degree relative with breast cancer also increases the risk for prostate cancer (RR 1.2-1.3)[494,525], however, this has not been confirmed by others[47]. Possibly these differences are due to different frequencies of BRCA1 or BRCA2 mutations (see below) in the families included in these studies.

Hereditary prostate cancer

Based on the study of families with a striking clustering of prostate cancer, it has been proposed that a subset of families exists with a strong hereditary predisposition to develop his tumour[526,527]. The name hereditary prostate cancer (HPC) has been coined to refer to this condition[526]. The HPC gene is postulated to be responsible for 43 % of all prostate cancer cases diagnosed before the age of 65 and for 9 % of cases before the age of 86[526]. Its predisposing alleles would confer a cumulative risk of developing prostate cancer of approximately 90 % at age 86[526,528], although a lifetime penetrance of 63 % has been suggested by another segregation analysis[529]. HPC gene loci have been mapped to chromosome 1 (HPC1 locus) [530] and to the X-chromosome[531], and await further identification.

Natural history of familial and hereditary prostate cancer

Kupelian *et al.*[532,533] as well as Rodriguez *et al.*[534] reported that familial prostate cancer, defined as a patient plus a first-degree affected relative, generally has a more aggressive course than non-familial prostate cancer. In contrast, Hanus *et al.* did not observe any such difference[535,536]. The genetic heterogeneity in the groups of patients included in these studies is probably larger than in the group fulfilling the more stringent criteria of hereditary prostate cancer. Grönberg *et al.*[536] compared sporadic, familial and hereditary prostate cancer cases and could not detect any differences in survival between them.

Some clinical/pathological parameters may nevertheless be different in HPC families: relatively early age of onset and lower Gleason scores have been documented[537-540]. In the subset of HPC families linked to the HPC1 locus on chromosome 1, younger age at diagnosis was confirmed, but higher-grade tumours and more advanced-stage disease were also observed[541]. Possibly different genetic subsets of HPC are associated with different clinical appearance and behaviour.

Prostate cancer and hereditary breast-ovarian cancer

Increased prostate cancer risks are also found in hereditary breast (-ovarian) Cancer families, associated with BRCA1 germline mutations (RR of 3, not typically early-onset types)[23]. Germline BRCA1 mutations may also be present in prostate cancer patients without a strong family history of breast and or ovarian cancer[542]. Prostate cancer has also been reported in families with mutant BRCA2 genes [543], however, whether prostate cancer risk is significantly increased in males with mutant BRCA2 genes is presently unknown.

Preventive options

Several screening protocols have been devised for men with a strong family history of prostate cancer. Generally, these include annual to biannual screening for prostate cancer, starting at the age of 40-50 with determination of serum PSA levels, digital rectal examination, followed by ultrasound studies and biopsies in the case of abnormal results [509,512,513,537,541,544-548]. It is presently unknown whether this type of screening is actually beneficial to this specific population. This also applies to male carriers of BRCA1 mutations[172]. Chemoprevention of prostate cancer by the use of finasteride (a 5α-reductase inhibitor) is presently being studied[549,550].

2.3.12 Skin, melanoma

Relative risks

Approximately 5-10 % of melanoma cases are familial and a family history of melanoma is an established risk factor for the development of this tumour[551,552]. Some of the other known risk factors, as for example pigment traits, have a genetic background and this may also be true for the number of melanocytic nevi[551,553,554]. These risk factors may contribute to melanoma clustering in families[552,555]. However, the study by Ford *et al*[556] failed to show a relationship between family history of melanoma and number of nevi, pigment traits and freckling. First-degree relatives of melanoma patients have an RR of approximately 2 to develop this tumour[49,556,557] and this is even higher in first-degree relatives of cases diagnosed before the age of 50 (RR 6.5)[49].

Hereditary melanoma

Melanoma and multiple atypical nevi are characteristic findings in the familial atypical multiple mole melanoma syndrome (FAMMM, also called familial dysplastic nevus syndrome), an autosomal dominant disorder which is genetically heterogeneous[557-560]. Although dysplastic nevi and melanomas have been reported to cluster within the same families and patients[561], familial melanoma may occur in the absence of dysplastic nevi. Including dysplastic nevi as an indicator for hereditary melanoma has confused genetic analysis[557,562]. It has been suggested to drop the term 'dysplastic nevus' altogether and redefine this lesion, naming it 'nevus with architectural disorder'[563].

Two genes associated with FAMMM, CDKN2A and (probably only in rare cases) CDK4, have been isolated so far[562,564,565]. Other FAMMM genes, *e.g.* on chromosome 1p36, remain to be identified. Germline CDKN2A mutations have been detected in approximately one fourth of melanoma-prone kindreds (two or more affected first-degree relatives) from the USA, Australia and Europe[566]. The cumulative risk to develop melanomas in FAMMM gene carriers have been estimated at approximately 50 % at age 80[567]. At age 40 it is 20-70 %, depending on the birth cohort. Gene carriers born after 1940 have been attributed the highest risk[568], although the calculations have been questioned[569].

An increased risk to develop melanomas also been documented in patients with xeroderma pigmentosum(XP)[422], hereditary retinoblastoma[570], oculocutaneous albinism[571] and Werner syndrome[572].

Preventive options

Periodical dermatological examination in patients with FAMMM and their first-degree relatives is recommended and the same applies to XP patients [563]. Although a sun-protective regiment (including topical sunscreens) may help to reduce the risk of melanomas and is therefore advised[563], the occurrence in FAMMM patients of melanomas in body parts not regularly exposed to sunlight, will obviously not be influenced by this strategy. Oral administration of isotretinoin has been used experimentally as chemoprevention for skin cancer in XP patients, but it is unclear whether this reduces melanoma risk in these patients, while serious side-effects (including teratogenic ones) exist [573,574].

2.3.13 Skin, non-melanoma

Relative risks

The influence of a family history of skin cancer on basal cell skin cancer risk is controversial. Two studies could not demonstrate a familial risk factor[575,576]. However, Wallberg *et al.*[577] found a history of skin cancer (in general) among siblings and/or parents to be the strongest (RR 10.9) of twelve tested potential risk factors. The authors could not explain this risk by correcting for hereditary pigmentary characteristics, as for example skin type, eye and hair colour, which are known to be associated with melanoma as well as non-melanoma skin cancer risk[578]. Hogan *et al.*[579] also found that a family history of skin cancer was associated with an increased risk to develop basal cell skin cancer (RR 1.22). In their series of 538 patients and matched controls, they also observed a significantly lower age at diagnosis in patients with a family history of skin cancer compared with those who did not. For squamous cell skin cancer, a positive family history of skin cancer (in general) may also increase risk, as has been observed by Hogan *et al.*[580] and Gamble *et al.*[581] (RR 2.3).

Skin cancer in hereditary disorders

Basal cell cancer of the skin is associated with a number of hereditary disorders, including basal cell nevus syndrome (Gorlin syndrome)[7], xeroderma pigmentosum[422], oculocutaneous albinism[571], ectodermal dysplasia (including Rothmund-Thomson syndrome[582]), Bazex syndrome[583], ataxia telangiectasia[467], Bloom syndrome[201] and Rombo syndrome[584].

Squamous cell carcinomas of the skin have also been found in several hereditary conditions, including Bloom syndrome (although less frequently than basal cell carcinomas)[201], Huriez syndrome[585], congenital dyskeratosis[586], ectodermal dysplasia (including keratitis-ichthyosis-deafness (KID) syndrome[587,588], multiple self-healing squamous epitheliomata (ESS1)[589], epidermolysis bullosa (dystrophic[590] as well as generalized atrophic benign type[591]), porokeratosis of Mibelli[592] and oculocutaneous albinism[571].

Sebaceous carcinomas, adenomas, epitheliomas and keratoacanthomas are typical for Muir-Torre syndrome[593](an HNPCC variant). Cutaneous cylindromas, which may show malignant transformation are found in familial (cutaneous) cylindromatosis[594,595]. A number of genodermatoses feature benign skin tumours, including Cowden syndrome (trichilemmomas)[596], neurofibromatosis type 1 (neurofibromas)[597], tuberous sclerosis (angiofibromas)[598] and blue rubber bleb nevus syndrome (haemangioma)[599].

Preventive options

Screening for skin cancer by self-examination and skin inspection by the physician is regularly performed in a number of the above mentioned disorders, including xeroderma pigmentosum and basal cell nevus syndrome and seem appropriate for all hereditary disorders with a significant increase in skin cancer risk. However, the precise clinical impact of screening for skin cancer and the preventive use of sun blocks in individuals with a hereditary predisposition is as yet unknown. As mentioned in section 2.3.12, oral administration of isotretinoin has been used experimentally as chemoprevention for skin cancer in XP patients[573,574], but serious side-effects (including teratogenic effects) exist. Oral isotretinoin as well as oral retinol have been reported to have no protective effect on subjects who are at a high risk to develop additional skin cancer because of a prior history of multiple skin cancer[600].

2.3.14 Stomach

Relative risks

A family history of stomach cancer is present in approximately 10-15 % of stomach cancer cases[601-603] and is an established risk factor for this tumour[604]. Depending on the number and degree of affected relatives, RR's have been estimated ranging from 1.5 (one affected first-degree relative) to 7.0 (unspecified family history of diffuse type stomach cancer)[601,605-608]. An association has been suggested between a family history of stomach cancer and increased risk of advanced stomach cancer (RR 2.6)[609] as well as multifocal stomach cancer (RR 2.1 in case of 1 first-degree affected relative, and RR 5.1 in case of more than 1 affected first-degree relatives)[610]. The risk of developing stomach cancer is increased in first-degree relatives of patients with Ewing's sarcoma (RR 2)[612], breast cancer (RR 1.2)[494], ovarian cancer (RR 3.7)[240] and brain/central nervous system cancer (RR 1.9)[49].

Helicobacter pylori infection is associated with stomach cancer[604]. Seropositivity for this microorganism has been found in association with a family history of stomach ulcers and stomach cancer[611]. Genetic factors may influence susceptibility to Helicobacter pylori infection and thus indirectly stomach cancer risk.

Hereditary gastric cancer

Hereditary diffuse type stomach cancer, associated with germline mutations in the E-cadherin gene has been observed in New Zealand's Maori families[613] as well as in European/Caucasian families[614,615]. As much as 8 % of the total stomach cancer burden has been attributed to Familial Adenomatous Polyposis (FAP)[616]. However, contrary to Japanese and Korean FAP patients, stomach cancer risk is not significantly increased in Western-European FAP patients[10], which might be due to differences in environmental (dietary) risk factors. Stomach cancer is part of hereditary non-polyposis colorectal cancer (HNPCC)[247,334,617], especially in the older generations.

Microsatellite instability, a common tumour feature of HNPCC (see 3.4.), has been detected more frequently in familial stomach cancer than in sporadic cases[618-620], suggesting the possibility of germline mutations in mismatch repair genes. However, not all studies support this association[621].

Stomach cancer has also been reported in patients with ataxia telangiectasia[467], Peutz-Jeghers syndrome[195], juvenile polyposis[498], Bloom syndrome[201], Fanconi anaemia[202], common variable immune deficiency[622], IgA deficiency[623].

Germline mutations in the hereditary breast cancer gene BRCA2 (see section 2.3.2.) may be associated with stomach cancer[97]. A rare case of familial gastric lymphoma has also been reported[624]. In general, gastric non-Hodgkin lymphoma is more likely to occur in individuals with a family history of gastric cancer (RR 18) which hints at shared etiological factors (*e.g.* H.pylori infection ?)[625]

Surveillance

Periodic gastroscopy and checking for (and treatment of) Helicobacter pylori infection could be considered in families with a strong history of gastric cancer, although its preventive effect in these families is as yet unknown. In HNPCC families with a history of stomach cancer, gastroscopy is recommended every 1-2 years, starting at the age of 30-35[335]. Upper gastrointestinal endoscopy is advised in FAP patients every three years, starting at age 25[10].

However, in Western-European patients this is mainly performed because of the risk of duodenal cancer. Upper gastrointestinal endoscopy is also recommended every 3-5 years in patients with Juvenile Polyposis[317] and every two years in patients with Peutz-Jeghers syndrome[205]. The benefit of screening for stomach cancer in these disorders has yet to be proven.

Table 1. Human cancer syndromes and selected other hereditary disorders with increased cancer risk

Disorder [synonym] (inheritance)[references]	Gene(s)	Tumours	Additional features & remarks
Ataxia Telangiectasia [Louis-Bar syndrome] (AR)[467,649,650]	ATM	HD, NHL, leukaemia (ALL, CLL), cancer of the breast, stomach, liver, larynx, ovary, parotid gland, skin, thyroid, astrocytoma.	cerebellar ataxia, oculocutaneous telangiectasia, skin hyper (café au lait spots) / hypopigmentation, immunodeficiency, increased serum AFP, increased radiosensitivity, increased chromosomal breakage
Bannayan-Riley-Ruvalcaba syndrome (AD)[651,652]	PTEN	hamartous polyps in ileum and colon, lipoma, haemangioma	high birth weight, hypotonia, gross motor delay, mental deficiency, epilepsy, lipid storage myopathy of proximal muscles, macrocephaly, facial/oral papules, prominent corneal nerves, pigmented macules on glans and shaft of penis, vulvar lentigines, joint hyperextensibility, pectus excavatum, scoliosis, variant of Cowden disease.
Basal Cell Nevus syndrome [Gorlin syndrome] (AD)[653]	PTCH	basal cell skin cancer, ovarian and cardiac fibroma, medulloblastoma	palmo/plantar pits, calcification of falx cerebri, macrocephaly, jaw cysts, frontoparietal bossing, broad nasal bridge, high arched palate, epidermal cysts, rib anomalies, vertebral anomalies, pectus excavatum/carinatum, prognathism, sloping shoulders, increased skin radiosensitivity
Bazex's syndrome (XL)[583]	Xq24-27	basal cell skin cancer	follicular atrophoderma ('ice pick marks'), hypotrichosis, milia, facial hyperpigmentation, hair shaft dystrophy

Beckwith-Wiedemann syndrome (spor., AD)[654-656]	11p15, p57kip2,	Wilms tumour, hepatoblastoma, adrenal cancer, pancreatoblastoma	macroglossia, gigantism, omphalocele, umbilical hernia hemihypertrophy, ear-lobe grooves, circular indentations on posterior rim of helix, craniofacial dysmorphisms, flame nevus, transitory hypoglycaemia, congenital heart defects, skeletal anomalies
Bloom syndrome (AR)[201,657]	BLM	NHL, basal cell skin cancer, ALL, AML, cancer of the cervix, breast, larynx, oropharynx, stomach	immunodeficiency, deficient growth, UV hypersensitivity, skin hyperpigmentation [including cafe au lait spots], hypopigmentation, long small facies with prominent nose and small mandible, intelligence varying from normal to severely retarded
Blue Rubber Bleb Nevus syndrome (AD)[599]	?	gastrointestinal polypoid haemangiomas, haemangiomatous cutaneous nevi = 'blue rubber blebs', haemangiomas in : liver, lungs, spleen, subcutaneous tissue, joint capsules	
Carney complex (AD)[658-660]	2p16, ?	psammomatous melanotic schwannoma, myxoma of breast, skin and heart, breast ductal adenoma, Sertoli or Leydig cell tumour testis, thyroid cancer, uterine leiomyoma	spotty pigmentation (lentiginosis) on skin and mucosa, pigmented nodular adrenocortical disease (Cushing disease)
Common Variable Immunodeficiency [Common Variable Hypogammaglobulinaemia, Late-onset Immunoglobulin deficiency] (AR,AD?,XL?,spor)[622,661]	?	NHL, stomach cancer, CLL	hypogammaglobulinaemia, immunodeficiency

Congenital Dyskeratosis [Zinsser-Engman-Cole syndrome, Dyskeratosis Congenita] (XL,AD,AR)[203,662]	DKC1, ?	squamous cell cancer of oro/nasopharynx, oesophagus, cervix and skin	bone marrow failure, mucosal leucoplakia, reticulated skin pigmentation, nail dystrophy, blepharitis, developmental delay, hyperhidrosis, hyperkeratosis, this entry summarises a group of disorders.
Cowden syndrome [Multiple hamartoma syndrome, incl. Lhermitte-Duclos disease] (AD)[596,663]	PTEN	multiple facial trichilemmomas, cancer and hamartomas of the breast, gastrointestinal and urinary tract hamartomas, lipomas, non-medullary thyroid cancer, ovarian tumor, uterine leiomyomas, skin haemangiomas	oral papules, macrocephaly, palmoplantar hyperkeratosis and pits, fibrocystic breast disease
Cystic Fibrosis (AR)[664-666]	CFTR	digestive tract cancer (ileal adenocarcinoma, pancreatic adenocarcinoma)	pancreatic fibrosis, lung emphysema, vas deferens aplasia, biliary cirrhosis, meconium ileus
Denys-Drash syndrome (de novo)[667]	WT1	Wilms tumour, gonadoblastoma	ambiguous genitals, nephropathy
Ectodermal dysplasia (XL,AD,AR)[668]	*	cancer of the cervix and skin (basal, squamous and Merckel cell type)	anomalies of hair, teeth, nails and skin (including sweat glands), this entry summarises a group of disorders
Epidermolysis Bullosa, Dystrophic Recessive [Hallopeau-Siemens type DEB, RDEB] (AR)[590]	COL7A1	squamous cell skin cancer, oesophageal cancer, oropharyngeal cancer	blistering of skin and mucosa (eyes, oropharynx, oesophagus, anus)
Epidermolysis Bullosa, Dystrophic Dominant [Passini type DEB, DDEB][669,670]	COL7A1	squamous cell skin cancer	blistering of skin and intraoral mucosa
Epidermolysis Bullosa, Generalised Atrophic Benign [GABEB][591,671](AR)	COL17A1	squamous cell skin cancer	skin blistering, dystrophic nails, enamel defects

Familial Adenomatous Polyposis [incl. Attenuated FAP, Gardner syndrome and Hereditary Desmoid disease] (AD)[10]	APC	gastrointestinal adenomatous polyps and cancer, desmoids, osteomas, hepatoblastoma, brain tumours, papillary thyroid cancer	congenital hyperplasia of the retinal pigment epithelium (CHRPE), dental abnormalities, sebaceous cysts, epidermoid cysts
Familial Atypical Multiple Mole Melanoma syndrome (Familial Dysplastic Nevus syndrome) (AD)[672]	CDKN2A, CDK4, 1p36	cutaneous malignant melanoma, pancreatic cancer, uveal melanoma	atypical/dysplastic nevi
Familial Cylindromatosis (AD)[595]	16q12-13	skin (malignant) cylindromas, trichoepithelioma, eccrine spiroadenomas	
Familial Gastrointestinal Stromal Tumours (AD)[673]	KIT	gastrointestinal stromal tumours,	perineal skin hyperpigmentation
Familial Giant Hyperplastic Polyposis (AD)[674]	?	hyperplastic polyps, adenomas and carcinoma of the colon and rectum	status as separate hereditary disorder unclear
Familial Non-Medullary Thyroid Cancer [FNMTC, incl. Familial Thyroid Tumours with Cell Oxyphilia] (AD ?)[675-677]	9p13.2, ?	non-medullary thyroid cancer	multinodular goitre
Familial non-VHL non-papillary clear-cell renal cancer (AD)[346]	?	non-papillary clear-cell renal cancer	
Familial Wilms tumour (AD)[362]	WT1, 17q12-q21, 19q13	Wilms tumour	

Fanconi Anaemia (AR)[202]	FANCA, FANCC, FANCG, 3p22, ?	AML, hepatocellular adenomas and cancer, oropharyngeal cancer, oesophageal cancer, gastric cancer, breast cancer, vulvar and cervical cancer, astrocytoma, medulloblastoma	pancytopenia, myelodysplastic syndrome, persistent fetal haemoglobin, increased chromosomal breakage, mental retardation, growth retardation, microcephaly, skeletal (radius) malformations, renal and ocular anomalies, cafe au lait spots
Fibrocystic Pulmonary Dysplasia (AD)[406,678]	?	lung cancer	lung fibrosis, this condition is possibly identical to Idiopathic Pulmonary Fibrosis.
Frasier syndrome (spor)	WT1	ovarian gonadoblastoma	ambiguous external genitals in 46,XY individuals
Hereditary Breast-Ovarian Cancer (AD)[679,680]	BRCA1, BRCA2, ?	breast, ovarian, prostate, colon and pancreatic cancer	
Hereditary Gastric Cancer (AD)[613]	ECAD	diffuse type gastric cancer	
Hereditary Mixed Polyposis syndrome(AD)[681]	6q	hamartomas,adenomas, hyperplastic polyps and cancer of the large bowel. Status as separate hereditary disorder unclear	
Hereditary Non-Polyposis Colorectal Cancer [HNPCC, Lynch syndrome] (AD)[682]	hMLH1, hMSH2, hPMS1, hPMS2, hMSH6, ?	colorectal, endometrial, ovarian, gastric, duodenal, renal pyelum/ureter and hepatobiliary tract cancer, glioma	
Hereditary Pancreatitis (AD)[683]	TRYP1, ?	pancreatic cancer	pancreatitis
Hereditary Papillary Renal Cell Carcinoma (AD)[350]	MET	papillary renal cell cancer	
Hereditary Prostate Cancer (AD)[537]	1q24-25, Xq27-28	prostate cancer	

Hereditary Retinoblastoma (AD)[684]	Rb1	retinoblastoma, osteosarcoma, soft tissue sarcoma, melanoma, lipoma	
Huriez syndrome (AD)[685]	4q28-31	squamous cell skin cancer	scleroatrophic and keratotic dermatosis of the limbs, hypoplastic nails, although an increased bowel cancer risk has been suggested, this is controversial[685]
Hyperparathyroidism-jaw tumour syndrome(AD)[355,686]	1q21-31	parathyroid adenoma/cancer, fibro-osseous maxillar/mandibular tumours, renal hamartomas	hyperparathyroidism, renal cysts, there is an overlap with Familial Isolated Hyperparathyroidism
Idiopathic Pulmonary Fibrosis [Hamann-Rich syndrome] (AD)[405,406]	?	lung cancer	lung fibrosis, possibly identical to Fibrocystic Pulmonary Dysplasia
Immunoglobulin A deficiency (AD, ?)[623,687,688]	?	gastric cancer	selective IgA deficiency, immunodeficiency
Juvenile Polyposis (AD)[317]	SMAD4 PTEN	gastrointestinal hamartous polyps, colorectal, gastric, small bowel and pancreatic cancer	wide range, including: pulmonary AV-fistula, macrocephaly, cleft lip/palate, urogenital abnormalities, congenital heart disease, malrotation of the gut; heterogenous group of disorders

Keratitis-Ichthyosis-Deafness syndrome [KID] (AD)[420]	?	squamous cell skin cancer, oropharyngeal (tongue) cancer	erythrokeratoderma, vascularising keratitis, palmoplantar hyperkeratosis, alopecia, hypohidrosis, neurosensory deafness
Li-Fraumeni syndrome [LFS, Sarcoma-Breast-Leukemia-Adrenal cancer syndrome, SBLA] (AD)[402,403]	TP53	soft-tissue and bone sarcomas, breast cancer, brain tumours (mainly astrocytoma), lung, adrenocortical cancer, ALL, HD	
Muir-Torre syndrome (AD)[593,689]	hMSH2, hMLH1, ?	sebaceous adenoma / epithelioma/ carcinoma, keratoacanthoma, internal malignancies including colorectal cancer	variant of HNPCC
Multiple Endocrine Neoplasia, type 1 [MEN1, Wermer's disease](AD)[502,690,691]	MEN1	pituitary adenomas, parathyroid adenoma, duodenal and gastric gastrinomas, pancreatic endocrine tumours, adrenal adenomas, carcinoids of thymus, bronchus and gastrointestinal tract, skin lipomas, angiofibromas and collagenomas	hyperparathyroidism
Multiple Endocrine Neoplasia, type 2A [MEN2A, Sipple disease, including Familial Medullary Thyroid cancer, FMTC] (AD)[692]	RET	medullary thyroid cancer, parathyroid adenoma, pheochromocytoma	hyperparathyroidism, in some cases: Hirschsprung disease, cutaneous lichen amyloidosis; in FMTC, risk of pheochromocytoma and parathyroid pathology is minimal.
Multiple Endocrine Neoplasia, type 2B [Mucosal Neuroma syndrome, Wagemann-Froboese syndrome] (AD, de novo)[693]	RET	medullary thyroid cancer, pheochromocytoma, gastrointestinal neurinomas	Marfanoid habitus, prominent corneal nerves
Multiple Self Healing Squamous Epitheliomata [Ferguson-Smith syndrome, ESS1] (AD)[589,694]	9q31	squamous cell skin cancer (self healing)	increased X-ray sensitivity of the skin

Neurofibromatosis, type 1 [NF1, von Recklinghausen disease] (AD, de novo)[695-699]	NF1	cutaneous neurofibromas, plexiform neurofibromas, optic gliomas, malignant peripheral nerve sheath tumours, juvenile chronic myelogenous leukaemia, gastrointestinal neurofibromas, phaeochromocytoma, (duodenal) carcinoid, rhabdomyosarcoma	café au lait spots skin, axillary freckling, Lisch nodules iris, macrocephaly, mental retardation, scoliosis, epilepsy
Neurofibromatosis, type 2 [NF2, Central Neurofibromatosis, Bilateral Acoustic Neurofibromatosis] (AD, de novo)[697]	NF2	vestibular schwannomas, other cranial nerve and cutaneous schwannomas, meningiomas, brain glioma/astrocytoma	congenital/juvenile cataract, café au lait spots skin
Oculocutaneous Albinism (AR)[571]	OCA1, OCA2	basal and squamous cell skin cancer, cutaneous malignant melanoma	albinism, nystagmus, visual loss
Perlman syndrome (AR)[356,369]	?	Wilms tumour, renal hamartomas	nephroblastomatosis, renal dysplasia, fetal gigantism, polyhydramnion, fetal ascites, cryptorchism, macrocephaly, facial dysmorphisms
Peutz-Jeghers syndrome (AD)[195,700]	STK11	gastrointestinal hamartomas and cancer, ovarian sex cord tumours with annular tubules (SCAT), Sertoli-Leydig tumours testis, adenoma malignum cervix, breast cancer	pigmentation of lips, oral mucosa, palmar, fingers and plantar areas
Porokeratosis of Mibelli (AD)[592]	?	squamous cell skin cancer	hyperkeratotic skin patches with central atrophy (crater-like)
Punctate Palmoplantar Keratoderma [Buschke-Fischer-Brauer disease] (AD ?)[701,702]	?	colorectal cancer	keratoderma, status as separate (hereditary) disorder unclear
Rombo syndrome (AD)[584]	?	basal cell skin cancer, trichoepithelioma	vermiculate atrophoderma, hypotrichosis, milia, peripheral vasodilatation with cyanosis

Rothmund-Thomson syndrome [Congenital Poikiloderma] (AR)[582,703]	8(?)	basal cell skin cancer, malignant eccrine poroma, osteosarcoma	poikilodermatosis, short stature, sparse/absent eye brows or lashes, juvenile cataract, defective dentition, dystrophic nails, radio-ulnar hypoplasia, increased X-ray sensitivity
Simpson-Golabi-Behmel syndrome (XL)[655,704]	GPC3	Wilms tumour, neuroblastoma	overgrowth, mental deficiency, hypotonia, vertebral anomalies, facial dysmorphisms
Sotos syndrome [Cerebral Gigantism] (spor,AD,AR ?)[655]	?	Wilms tumour	overgrowth, advanced bone age, delayed mental development, macrodolichocephaly, frontal bossing, ocular hypertelorism, prominent mandible
Tuberous Sclerosis [Bourneville-Pringle disease] (AD, de novo)[598.705]	TSC1,TSC2	facial angiofibromas, ungual fibromas, cardiac rhabdomyomas, gingival fibromas, brain subependymal nodules, retinal hamartomas, renal and liver angiomyolipomas, renal cell cancer, rectal adenomatous polyps, colorectal hamartomas, giant cell astrocytoma	intracranial (periventricular) calcifications, dental pits, renal cysts, bone cysts, hypomelanotic (ashleaf) macules, shagreen patches, facial fibrous plaques, epilepsy (West syndrome)
Turcot syndrome [Brain Tumour-Colorectal Polyp(osis) syndrome] (AR ? AD ?)[706,707]	?	gastrointestinal polyposis and brain tumours (mainly astrocytoma and medulloblastoma).	café au lait spots status as separate hereditary disorder unclear: shows overlap with both HNPCC and Familial Adenomatous Polyposis.
von Hippel-Lindau disease (AD)[708]	VHL	central nervous system and retinal haemangioblastomas, non-papillary clear cell renal cancer, pheochromocytoma, endocrine pancreatic tumours, endolymphatic sac tumours,	renal, hepatic, pancreatic, lung adnexal and epididymal cyst(adenoma)s

WAGR (de novo)[709]	c.g.d.: WT1 + PAX6 + ?	Wilms tumour, gonadoblastoma	aniridia, mental retardation, genitourinary anomalies
Werner syndrome (AR)[710]	WRN	soft tissue sarcoma, meningioma, AML, cutaneous malignant melanoma, non-medullary thyroid cancer	premature aging, short stature, slender limbs, stocky trunk, beaked nose, scleroderma-like skin changes, arteriosclerosis, juvenile cataract, diabetes mellitus, MDS
Xeroderma Pigmentosum (AR)[711]	XPA, XPB, XPC, XPD, XPG, ?	basal and squamous cell skin cancer, cutaneous malignant melanoma, keratoacanthoma	extreme UV sensitivity, neurological degeneration and mental retardation
X-linked Agammaglobulinemia [Bruton type Agammaglobulinemia] (XL)[688,712,713]	BTK	HD, NHL, ALL, CML, thymoma, colorectal cancer	agammaglobulinemia, immunodeficiency
X-linked Lymphoproliferative syndrome [Duncan syndrome] (XL)[714]	SH2D1A	NHL (Burkitt lymphoma)	immunodeficiency (Epstein-Barr virus susceptibility) , aplastic anaemia

This table lists the hereditary cancer predisposition syndromes as well as other hereditary disorders associated with increased cancer susceptibility as mentioned in section 2.3 of the paper.
Inheritance: *AD = autosomal dominant; AR = autosomal recessive; de novo = de novo gene mutation(i.e. parents have normal, wild type, genotypes); spor= sporadic cases (i.e. single cases in a family; genetic background, e.g. de novo mutations ?, as yet unknown); XL = X-linked inheritance;*
? = inconclusive evidence with regard to inheritance pattern.
Tumours: *in case a range of tumours is listed, those which most frequently occur in the mentioned hereditary disorder are underlined. AML = acute myeloid leukaemia, ALL = acute lymphoblastic leukaemia, CML= chronic myeloid leukaemia, HD = Hodgkin disease, NHL = Non-Hodgkin lymphoma*
Gene(s): *multiple entries reflect the fact that mutations in different genes can cause this disorder; ..p or ..q = gene has been mapped to the short (p) or long (q) arm of this chromosome, but has not been isolated yet; c.g.d. = contiguous gene defect (several genes in a row have been deleted from a chromosomal region); ? = (additional) unknown (i.e. unmapped) gene(s);*
*(?) = gene location uncertain; * = genes/locations are too numerous to be listed*

2.4 Hereditary cancer registries

Registries of families with an inherited predisposition to cancer were established in several parts of the world since the early 1980s, although the oldest one, that of St. Marks Hospital in London, dates back to the 1920s . The primary reason for the establishment of these hereditary cancer registries was the experience that the possible hereditary nature of familial clustering of cancer was largely neglected by the medical community and that genealogic studies of suspected families were not or incompletely performed. Consequently, only a small proportion of the high risk individuals were under surveillance[626,627]. The main objectives of the registries were (1) to promote the identification of family members at increased risk for cancer, (2) to enhance the (continuous) participation of these relatives in early detection programs and (3) to promote research and education in these relatively rare hereditary cancer syndromes[628].

There are different types of hereditary cancer registries: some are on a national basis, others operate regionally. Registries may focus on families with specific human cancer syndromes as familial adenomatous polyposis and/or hereditary non-polyposis colorectal cancer (*e.g.* the St Marks Polyposis Registry and the Danish Polyposis/HNPCC Registry)[629,630], Von Hippel Lindau disease[373] and familial or hereditary ovarian cancer[631,632], or may collect data on a wide spectrum of hereditary cancers (the Netherlands Hereditary Cancer Registry)[294]. Most registries include genetic field workers who assist the clinical specialists with the pedigree studies. These centres usually have a computerized follow up system to ensure the continuity of surveillance. In a few centres (St Marks Hospital, London; Johns Hopkins University Hospital , Baltimore) also the clinical management is centralized. In other regions and countries these clinical services (especially diagnosis, counselling and tumour screening) are provided by cancer family clinics[633-636] or by different independent departments of academic and regional hospitals and universities.

Several reports have described a considerable reduction of morbidity and mortality in families monitored by a central registry[294,629,637,638]. Due to the increased awareness of hereditary cancer by physicians and the general public, more and more families are being identified. These families include not only those with a pattern of inheritance that is definitely dominant, but also families with a pattern of inheritance that is less well defined. Thus, an increasing number of individuals is

under surveillance. This raises questions with respect to the effectiveness of the recommended screening protocols are cost-benefit ratios. Registries may help to answer these questions in the years to come.

2.5 Summary and conclusions

Although knowledge on the molecular aspects of hereditary cancer predisposition is rapidly accumulating, data on the natural history of the human cancer syndromes are still very incomplete. The same applies to the impact of cancer preventive options offered to families with these syndromes. This does not come as a surprise, because some of these syndromes are very rare and it has been difficult in the past to identify with certainty those syndromes which are characterized by common type cancers. The patients and relatives included in the studies on risks and natural history associated with a family history of cancer will often have been very heterogeneous with respect to the presence, absence and type of inherited cancer predisposition and impact of preventive options. Increasingly, molecular tools are being made available to help identifying this predisposition. Only now we are becoming able to study cohorts of patients with a proven inherited predisposition for common types of cancer for the natural history of these disorders and the clinical impact of various cancer prevention strategies[639].

How much effort should we put in trying to diagnose hereditary cancer predisposition in the clinic, outside research settings ? On the one hand, the preventive options for almost all of the discussed hereditary cancer predisposition disorders are still of unproven benefit[640,641]. In addition, diagnosing these genetic disorders may have adverse psychological effects[642] and may lead to genetic discrimination[643-645].

On the other hand, patients with possible hereditary cancer predisposition and their relatives may have a strong wish to know the nature of the disorder in their family and may want to give prevention programs the benefit of the doubt. Some will state that not knowing whether they are at increased risk to develop cancer in itself causes them psychological harm, and that they want to do whatever possible to decrease their risk of cancer. In addition, knowledge on the genetics of the disorder may be of importance in their family planning. Of course, in some disorders, *e.g.* Multiple Endocrine Neoplasia 2A and Familial Adenomatous Polyposis, benefits of genetic testing are clinically more obvious.

Clearly, genetic counselling[646] and an informed consent procedure[647] will have to precede genetic testing and participation in cancer prevention programs. In our opinion, the majority of testing and prevention will need to be performed in a research setting until cancer risk prediction will become more precise and effects of preventive measures and psychosocial consequences will be clarified. Given the growing market for commercial DNA testing, and the fact that physicians ordering the tests are not necessarily trained in the interpretation of test results and in genetic counselling[648], these issues should be closely monitored by the professional community.

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2.7 References

1 Ponder BAJ. Inherited predisposition to cancer. *Trends Genet* 1990;**7**:213-218.

2 Easton DF. The inherited component of cancer. *Br Med Bull* 1994;**50**:527-535.

3 Lynch HT, Lynch JF. Breast cancer genetics in an oncology clinic: 328 consecutive patients. *Cancer Genet Cytogenet* 1986;**22**:369-371.

4 Claus EB, Schildkraut JM, Thompson WD, Risch NJ. The genetic attributable risk of breast and ovarian cancer. *Cancer* 1996;**77**:2318-2324.

5 Griffin JP, Hughes GV, Peeling WB. A survey of the familial incidence of adenocarcinoma of the kidney. *Br J Urol* 1967;**39**:63-66.

6 Mclaughlin JK, Mandel JS, Blot WJ, Schuman LM, Mehl ES, Fraumeni JF, Jr. A population--based case--control study of renal cell carcinoma. *J Natl Cancer Inst* 1984;**72**:275-284.

7 Shanley S, Ratcliffe J, Hockey A, et al. Nevoid basal cell carcinoma syndrome: review of 118 affected individuals. *Am J Med Genet* 1994;**50**:282-290.

8 Gorlin RJ. Nevoid basal-cell carcinoma syndrome. *Medicine* 1987;**66**:98-113.

9 Vasen HFA, van der Feltz M, Raue F, et al. The natural course of multiple endocrine neoplasia type IIb. A study of 18 cases. *Arch Intern Med* 1992;**152**:1250-1252.

10 Phillips RKS, Spiegelman AD, Thomson JPS. *Familial Adenomatous Polyposis and other polyposis syndromes*. London: Edward Arnold, 1994;

11 Perera FP. Environment and cancer: Who are susceptible? *Science* 1997;**278**:1068-1073.

12 d'Errico A, Taioli E, Chen X, Vineis P. Genetic metabolic polymorphisms and the risk of cancer: A review of the literature. *Biomarkers* 1996;**1**:149-173.

13 Borch-Johnsen K, Olsen JH, Sorensen TIA. Genes and family environment in familial clustering of cancer. *Theor Med* 1994;**15**:377-386.

14 Love RR, Evans AM, Joston DM. The accuracy of patient reports of a family history of cancer. *J Chron Dis* 1985;**4**:289-293.

15 Carlson KM, Bracamontes J, Jackson CE, et al. Parent-of-origin effects in multiple endocrine neoplasia type 2B. *Am J Hum Genet* 1994;**55**:1076-1082.

16 Swift M, Morrell D, Massey RB, Chase CL. Incidence of cancer in 161 families affected by ataxia-teleangiectasia. *N Engl J Med* 1991;**325**:1831-1836.

17 Inskip HM, Kinlen LJ, Taylor AMR, Woods CG, Arlett CF. Risk of breast cancer and other cancers in heterozygotes for ataxia- telangiectasia. *Brit J Cancer* 1999;**79**:1304-1307.

18 Harrington DS, Weisenburger DD, Purtilo DT. Malignant lymphoma in the X-linked lymphoproliferative syndrome. *Cancer* 1987;**59**:1419-1429.

19 Vasen HFA, Taal BG, Griffioen G, et al. Clinical heterogeneity of familial colorectal cancer and its influence on screening protocols. *Gut* 1994;**35**:1262-1266.

20 Easton DF, Ford D, Bishop DT, et al. Breast and ovarian cancer incidence in BRCA1-mutation carriers. *Am J Hum Genet* 1995;**56**:265-271.

21 Marshall E. Search for a killer: focus shifts from fat to hormones. *Science* 1993;**259**:618-621.

22 Vasen HFA, Offerhaus GJA, den Hartog-Jager FCA, et al. The tumour spectrum in hereditary non-polyposis colorectal cancer: a study of 24 kindreds in The Netherlands. *Int J Cancer* 1990;**46**:31-34.

23 Ford D, Easton DF, Bishop DT, Narod SA, Goldgar DE. Risks of cancer in BRCA1-muation carriers. *Lancet* 1994;**343**:692-695.

24 Li FP, Fraumeni jr JF. Soft-tissue sarcomas, breast cancer, and other neoplsms: a familial syndrome? *Ann Intern Med* 1969;**71**:747-752.

25 Vasen HF, Nieuwenhuijzen Kruseman AC, Berkel H, et al. Multiple endocrine neoplasia syndrome type 2: the value of screening and central registration. A study of 15 kindreds in The Netherlands. *Am J Med* 1987;**83**:847-852.

26 Fraumeni jr JF, Devesa SS, Hoover RN, Kinlen LJ. Epidemiology of cancer. In: DeVita jr VT, Hellman S, Rosenberg SA, eds. *Cancer. Principles & practise of oncology*. Philadelphia: J.B.Lippincott Co. 1993;150-181.

27 Dupont WD, Plummer WD. Understanding the relationship between relative and absolute risk. *Cancer* 1996;**77**:2193-2199.

28 Lynch HT, Kimberling WJ, Lynch JF, Brennan K. Familial bladder cancer in an oncology clinic. *Cancer Genet Cytogenet* 1987;**27**:161-165.

29 Bravo MP, Del Rey-Calero J, Conde M. Risk factors of bladder cancer in Spain. *Neoplasma* 1987;**34**:633-637.

30 Lynch HT, Walzak MP, Fried R, Domina AH, Lynch JF. Familial factors in bladder carcinoma. *J Urol* 1979;**122**:458-461.

31 Purtilo DT, McCarthy B, Yang JP, Friedell GH. Familial urinary bladder cancer. *Semin Oncol* 1979;**6**:254-256.

32 Kiemeney LALM, Schoenberg M. Familial transitional cell carcinoma. *J Urol* 1996;**156**:867-872.

33 Kantor AF, Hartge P, Hoover RN, Fraumeni jr JF. Familial and environmental interactions in bladder cancer risk. *Int J Cancer* 1985;**35**:703-706.

34 Kramer AA, Graham S, Burnett WS, Nasca P. Familial aggregation of bladder cancer stratified by smoking status. *Epidemiology* 1991;**2**:145-148.

35 Rao KVN, Detrisac CJ, Steele VE, Hawk ET, Kelloff GJ, McCormick DL. Differential activity of aspirin, ketoprofen and sulindac as cancer chemopreventive agents in the mouse urinary bladder. *Carcinogenesis* 1996;**17**:1435-1438.

36 Feuer EJ, Wun LM, Boring CC, Flanders WD, Timmel MJ, Tong T. The lifetime risk of developing breast cancer. *J Natl Cancer Inst* 1993;**85**:892-897.

37 Burki N, Gencik A, Torhorst JKH, Weber W, Müller H. Familial and histological analyses of 138 breast cancer patients. *Breast Cancer Res Treat* 1987;**10**:159-167.

38 Demeter JG, Waterman NG, Verdi GD. Familial male breast carcinoma. *Cancer* 1990;**65**:2342-2343.

39 Kozak FK, Hall JG, Baird PA. Familial breast cancer in males: a case report and review of the literature. *Cancer* 1986;**58**:2736-2739.

40 Everson RB, Fraumeni jr JF, Wilson RE, et al. Familial male breast cancer. *Lancet* 1976;**january 3**:9-12.

41 Pharoah PDP, Day NE, Duffy S, Easton DF, Ponder BAJ. Family history and the risk of breast cancer: A systematic review and meta-analysis. *Int J Cancer* 1997;**71**:800-809.

42 Anderson DE, Badzioch MD. Breast cancer risks in relatives of male breast cancer patients. *J Natl Cancer Inst* 1992;**84**:1114-1117.

43 Olsson H, Andersson H, Johansson O, Möller TR, Kristoffersson U, Wenngren E. Population-based cohort investigations of the risk for malignant tumors in first-degree relatives and wives of men with breast cancer. *Cancer* 1993;**71**:1273-1278.

44 Rosenblatt KA, Thomas DB, McTiernan A, et al. Breast cancer in men: aspects of familial aggregation. *J Natl Cancer Inst* 1991;**83**:849-854.

45 Lenfant-Pejovic MH, Mlika-Cabanne N, Bouchardy C, Auquier A. Risk factors for male breast cancer: a franco-swiss case-control study. *Int J Cancer* 1990;**45**:661-665.

46 Anderson DE, Badzioch MD. Familial effects of prostate and other cancers on lifetime breast cancer risk. *Breast Cancer Res Treat* 1993;**28**:107-113.

47 Anderson DE, Badzioch MD. Familial breast cancer risks. Effects of prostate and other cancers. *Cancer* 1993;**72**:114-119.

48 Claus EB, Risch N, Thompson WD. The calculation of breast cancer risk for women with a first degree family history of ovarian cancer. *Breast Cancer Res Treat* 1993;**28**:115-120.

49 Goldgar DE, Easton DF, Cannon Albright LA, Skolnick MH. Systematic population-based assessment of cancer risk in first-degree relatives of cancer probands. *J Natl Cancer Inst* 1994;**86**:1600-1608.

50 McTiernan A, Gilligan MA, Redmond C. Assessing individual risk for breast cancer: Risky business. *J Clin Epidemiol* 1997;**50**:547-556.

51 Gail MH, Brinton LA, Byar DP, et al. Projecting individualized probabilities od developing breast cancer for white females who are being examined annualy. *J Natl Cancer Inst* 1989;**81**:1879-1886.

52 Bondy ML, Lustbader ED, Halabi S, Ross E, Vogel VG. Validation of a breast cancer risk assessment model in women with a positive family history. *J Natl Cancer Inst* 1994;**8**:620-625.

53 Spiegelman D, Colditz GA, Hunter D, Hertzmark E. Validation of the Gail et al. model for predicting individual breast cancer risk. *J Natl Cancer Inst* 1994;**8**:600-607.

54 Gail MH, Benichou J. Validation studies on a model for breast cancer risk. *J Natl Cancer Inst* 1994;**8**:573-575.

55 Benichou J, Gail MH, Mulvihill JJ. Graphs to estimate an individualized risk of breast cancer. *J Clin Oncol* 1996;**14**:103-110.

56 Claus EB, Thompson WD. Genetic analysis of breast cancer in the cancer and steroid hormone study. *Am J Hum Genet* 1991;**48**:232-242.

57 Claus EB, Risch N, Thompson WD. Autosomal dominant inheritance of early-onset breast cancer. *Cancer* 1993;**73**:643-651.

58 McGuigan KA, Ganz PA, Breant C. Agreement between breast cancer risk estimation methods. *J Natl Cancer Inst* 1996;**88**:1315-1317.

59 Becher H, ChangClaude J. Estimating disease risks for individuals with a given family history in different populations with an application to breast cancer. *Genet Epidemiol* 1996;**13**:229-242.

60 Schmidt S, Becher H, Chang-Claude J. Breast cancer risk assessment: use of complete pedigree information and the effect of misspecified ages at diagnosis of affected relatives. *Hum Genet* 1998;**102**:348-356.

61 Colditz GA, Rosner BA, Speizer FE. Risk factors for breast cancer according to family history of breast cancer. *J Natl Cancer Inst* 1996;**88**:365-371.

62 Andrieu N, Smith T, Duffy S, et al. The effects of interaction between familial and reproductive factors on breast cancer risk: a combined analysis of seven case-control studies. *Br J Cancer* 1998;**77**:1525-1536.

63 Egan KM, Stampfer MJ, Rosner BA, et al. Risk factors for breast cancer in women with a breast cancer family history. *Cancer Epidemiol Biomarkers Prev* 1998;**7**:359-364.

64 Katsouyanni K, Signorello LB, Lagiou P, Egan K, Trichopoulos D. Evidence that adult life risk factors influence the expression of familial propensity to breast cancer. *Epidemiology* 1997;**8**:592-595.

65 Ursin G, Henderson BE, Haile RW, et al. Does oral contraceptive use increase the risk of breast cancer in women with BRCA1/BRCA2 mutations more than in other women? *Cancer Res* 1997;**57**:3678-3681.

66 Dupont WD, Page DL, Parl FF, et al. Long-term risk of breast cancer in women with fibroadenoma. *N Engl J Med* 1994;**1**:10-15.

67 Skolnick MH, Cannon-Albright LA, Goldgar DE, et al. Inheritance of proliferative breast disease in breast cancer kindreds. *Science* 1990;**250**:1715-1720.

68 Brunet JS, Ghadirian P, Rebbeck TR, et al. Effect of smoking on breast cancer in carriers of mutant BRCA1 or BRCA2 genes. *J Natl Cancer Inst* 1998;**90**:761-766.

69 Miki Y, Swensen J, Shattuck-Eidens D, et al. A strong candidate for the breast and ovarian cancer susceptibility gene BRCA1. *Science* 1994;**266**:66-71.

70 Easton DF, Bishop DT, Ford D, Crockford GP, etal. Genetic linkage analysis in familial breast and ovarian cancer: Results from 214 families. *Am J Hum Genet* 1993;**52**:678-701.

71 Wooster R, Bignell G, Lancaster J, et al. Identification of the breast cancer susceptibility gene BRCA2. *Nature* 1995;**378**:789-792.

72 Tavtigian SV, Simard J, Rommens J, et al. The complete BRCA2 gene and mutations in chromosome 13q- linked kindreds. *Nat Genet* 1996;**12**:333-337.

73 Wooster R, Neuhausen SL, Mangion J, et al. Localization of a breast cancer susceptibility gene, BRCA2, to chromosome 13q12-13. *Science* 1994;**265**:2088-2090.

74 Phelan CM, Lancaster JM, Tonin P, et al. Mutation analysis of the BRCA2 gene in 49 site-specific breast cancer families. *Nat Genet* 1996;**13**:120-122.

75 Couch FJ, Deshano ML, Blackwood MA, et al. BRCA1 mutations in women attending clinics that evaluate the risk of breast cancer. *N Engl J Med* 1997;**336**:1409-1415.

76 Narod SA, Ford D, Devilee P, et al. An evaluation of genetic heterogeneity in 145 breast ovarian cancer families. *Am J Hum Genet* 1995;**56**:254-264.

77 Couch FJ, Farid LM, Deshano ML, et al. BRCA2 germline mutations in male breast cancer cases and breast cancer families. *Nat Genet* 1996;**13**:123-125.

78 Thorlacius S, Olafsdottir G, Tryggvadottir L, et al. A single BRCA2 mutation in male and female breast cancer families from Iceland with varied cancer phenotypes. *Nat Genet* 1996;**13**:117-119.

79 Berry DA, Parmigiani G, Sanchez J, Schildkraut J, Winer E. Probability of carrying a mutation of breast-ovarian cancer gene BRCA1 based on family history. *J Natl Cancer Inst* 1997;**89**:227-238.

80 Shattuck-Eidens D, Oliphant A, Mcclure M, et al. BRCA 1 sequence analysis in women at high risk for susceptibility mutations: Risk factor analysis and implications for genetic testing. *JAMA* 1997;**278**:1242-1250.

81 Fitzgerald MG, Macdonald DJ, Krainer M, et al. Germ-line BRCA1 mutations in Jewish and non-Jewish women with early-onset breast cancer. *N Engl J Med* 1996;**334**:143-149.

82 Langston AA, Malone KE, Thompson JD, Daling JR, Ostrander EA. BRCA1 mutations in a population-based sample of young women with breast cancer. *N Engl J Med* 1996;**334**:137-142.

83 Struewing JP, Tarone RE, Brody LC, Li FP, Boice JD. BRCA1 mutations in young women with breast cancer. *Lancet* 1996;**347**:1493

84 Krainer M, Silva-Arrieta S, Fitzgerald MG, et al. Differential contributions of BRCA1 and BRCA2 to early-onset breast cancer. *N Engl J Med* 1997;**336**:1416-1421.

85 Newman B, Mu H, Butler LM, Millikan RC, Moorman PG, King MC. Frequency of breast cancer attributable to BRCA1 in a population-based series of American women. *JAMA* 1998;**279**:915-921.

86 Malone KE, Daling JR, Thompson JD, O'Brien CA, Francisco LV, Ostrander EA. BRCA1 mutations and breast cancer in the general population: Analyses in women before age 35 years and in women before age 45 years with first-degree family history. *JAMA* 1998;**279**:922-929.

87 Ford D, Easton DF, Peto J. Estimates of the gene frequency of BRCA1 and its contribution to breast and ovarian cancer incidence. *Am J Hum Genet* 1995;**57**:1457-1462.

88 Simard J, Tonin P, Durocher F, et al. Common origins of BRCA1 mutations in Canadian breast and ovarian cancer families. *Nat Genet* 1994;**8**:392-398.

89 Egan KM, Newcomb PA, Longnecker MP, et al. Jewish religion and risk of breast cancer. *Lancet* 1996;**347**:1645-1646.

90 Offit K, Gilewski T, McGuire P, et al. Germline BRCA1 185delAG mutations in Jewish women with breast cancer. *Lancet* 1996;**347**:1643-1645.

91 Struewing JP, Abeliovich D, Peretz T, et al. The carrier frequency of the BRCA1 185delAG mutation is approximately 1 percent in Ashkenazi Jewish individuals (vol 11, pg 198, 1995). *Nat Genet* 1996;**12**:110

92 Neuhausen SL, Mazoyer S, Friedman L, et al. Haplotype and phenotype analysis of six recurrent BRCA1 mutations in 61 families: Results of an international study. *Am J Hum Genet* 1996;**58**:271-280.

93 Andersen TI, Borresen AL, Moller P. A common BRCA1 mutation in Norwegian breast and ovarian cancer families? *Am J Hum Genet* 1996;**59**:486-487.

94 Johansson O, Ostermeyer EA, Hakansson S, et al. Founding BRCA1 mutations in hereditary breast and ovarian cancer in southern Sweden. *Am J Hum Genet* 1996;**58**:441-450.

95 Neuhausen S, Gilewski T, Norton L, et al. Recurrent BRCA2 6174delT mutations in Ashkenazi Jewish women affected by breast cancer. *Nat Genet* 1996;**13**:126-128.

96 Johannesdottir G, Gudmundsson J, Bergthorsson JT, et al. High prevalence of the 999del5 mutation in Icelandic breast and ovarian cancer patients. *Cancer Res* 1996;**56**:3663-3665.

97 Berman DB, Costalas J, Schultz DC, Grana G, Daly M, Godwin AK. A common mutation in BRCA2 that predisposes to a variety of cancers is found in both Jewish Ashkenazi and non-Jewish individuals. *Cancer Res* 1996;**56**:3409-3414.

98 Couch FJ, Hartmann LC. BRCA1 testing - Advances and retreats. *JAMA* 1998;**279**:955-957.

99 Friedman LS, Gayther SA, Kurosaki T, et al. Mutation analysis of BRCA1 and BRCA2 in a male breast cancer population. *Am J Hum Genet* 1997;**60**:313-319.

100 Mavraki E, Gray IC, Bishop DT, Spurr NK. Germline BRCA2 mutations in men with breast cancer. *Br J Cancer* 1997;**76**:1428-1431.

101 Haraldsson K, Loman N, Zhang QX, Johannsson O, Olsson H, Borg A. BRCA2 germ-line mutations are frequent in male breast cancer patients without a family history of the disease. *Cancer Res* 1998;**58**:1367-1371.

102 Easton DF. Cancer risks in A-T heterozygotes. *Int J Radiat Biol* 1994;**66**:S177-S182.

103 Nowak R. Discovery of AT gene sparks biomedical bonanza. *Science* 1995;**268**:1700-1701.

104 Vorechovsky I, Luo LP, Lindblom A, et al. ATM mutations in cancer families. *Cancer Res* 1996;**56**:4130-4133.

105 Vorechovsky I, Rasio D, Luo LP, et al. The ATM gene and susceptibility to breast cancer: Analysis of 38 breast tumors reveals no evidence for mutation. *Cancer Res* 1996;**56**:2726-2732.

106 Athma P, Rappaport R, Swift M. Molecular genotyping shows that ataxia-telangiectasia heterozygotes are predisposed to breast cancer. *Cancer Genet Cytogenet* 1996;**92**:130-134.

107 Fitzgerald MG, Bean JM, Hegde SR, et al. Heterozygous ATM mutations do not contribute to early onset of breast cancer. *Nat Genet* 1997;**15**:307-310.

108 Bishop DT, Hopper J. AT-tributable risks? *Nat Genet* 1997;**15**:226

109 Werneke U. Ataxia telangiectasia and risk of breast cancer. *Lancet* 1997;**350**:739-740.

110 Swift M. Ataxia telangiectasia and risk of breast cancer. *Lancet* 1997;**350**:740

111 Chen JD, Giesler G, Birkholtz G, Lindblom P, Rubio C, Lindblom A. The role of ataxia-telangiectasia heterozygotes in familial breast cancer. *Cancer Res* 1998;**58**:1376-1379.

112 Bay JO, Grancho M, Pernin D, et al. No evidence for constitutional ATM mutation in breast/gastric cancer families. *Int J Oncol* 1998;**12**:1385-1390.

113 Sobol H, Birnbaum D, Eisinger F. Evidence for a third breast cancer susceptibility gene. *Lancet* 1994;**344**:1151-1152.

114 Kerangueven F, Essioux L, Dib A, et al. Loss of heterozygosity and linkage analysis in breast carcinoma: Indication for a putative third susceptibility gene on the short arm of chromosome 8. *Oncogene* 1995;**10**:1023-1026.

115 Seitz S, Rohde K, Bender E, et al. Strong indication for a breast cancer susceptibility gene on chromosome 8p12-p22: Linkage analysis in German breast cancer families. *Oncogene* 1997;**14**:741-743.

116 Rebbeck TR, Couch FJ, Kant J, et al. Genetic heterogeneity in hereditary breast cancer: Role of BRCA1 and BRCA2. *Am J Hum Genet* 1996;**59**:547-553.

117 Serova OM, Mazoyer S, Puget N, et al. Mutations in BRCA1 and BRCA2 in breast cancer families: Are there more breast cancer-susceptibility genes? *Am J Hum Genet* 1997;**60**:486-495.

118 Struewing JP, Hartge P, Wacholder S, et al. The risk of cancer associated with specific mutations of BRCA1 and BRCA2 among ashkenazi jews. *N Engl J Med* 1997;**336**:1401-1408.

119 Levy-Lahad E, Catane R, Eisenberg S, et al. Founder BRCA1 and BRCA2 mutations in Ashkenazi Jews in Israel: Frequency and differential penetrance in ovarian cancer and in breast- ovarian cancer families. *Am J Hum Genet* 1997;**60**:1059-1067.

120 Ford D, Easton DF, Stratton M, et al. Genetic heterogeneity and penetrance analysis of the BRCA1 and BRCA2 genes in breast cancer families. *Am J Hum Genet* 1998;**62**:676-689.

121 Schubert EK, Lee MK, Mefford HC, et al. BRCA2 in American families with four or more cases of breast or ovarian cancer: Recurrent and novel mutations, variable expression, penetrance, and the possibility of families whose cancer is not attributable to BRCA1 or BRCA2. *Am J Hum Genet* 1997;**60**:1031-1040.

122 Thorlacius S, Struewing JP, Hartge P, et al. Population-based study of risk of breast cancer in carriers of BRCA2 mutation. *Lancet* 1998;**352**:1337-1339.

123 Gayther SA, Warren W, Mazoyer S, et al. Germline mutations of the BRCA 1 gene in breast and ovarian cancer families provide evidence for a genotype- phenotype correlation. *Nat Genet* 1995;**11**:428-433.

124 Collins FS. BRCA1 - Lots of mutations, lots of dilemmas. *N Engl J Med* 1996;**334**:186-188.

125 Gayther SA, Mangion J, Russell P, et al. Variation of risks of breast and ovarian cancer associated with different germline mutations of the BRCA2 gene. *Nat Genet* 1997;**15**:103-105.

126 Neuhausen SL, Godwin AK, Gershoni-Baruch R, et al. Haplotype and phenotype analysis of nine recurrent BRCA2 mutations in 111 families: Results of an international study. *Am J Hum Genet* 1998;**62**:1381-1388.

127 Easton DF, Steele L, Fields P, et al. Cancer risks in two large breast cancer families linked to BRCA2 on chromosome 13q12-13. *Am J Hum Genet* 1997;**61**:120-128.

128 Lakhani SR, Easton DF, Stratton MR. Pathology of familial breast cancer:differences between breast cancers in carriers of BRCA1 or BRCA2 mutations and sporadic cases. *Lancet* 1997;**349**:1505-1510.

129 Marcus JN, Page DL, Watson P, Narod SA, Lenoir GM, Lynch HT. BRCA1 and BRCA2 hereditary breast carcinoma phenotypes. *Cancer* 1997;**80**:543-556.

130 Sobol A, Stoppa-Lyonnet D, Bressac-de Paillerets B, et al. Truncation at conserved terminal regions of BRCA1 protein is associated with highly proliferating hereditary breast cancers. *Cancer Res* 1996;**56**:3216-3219.

131 Lynch BJ, Holden JA, Buys SS, Neuhausen SL, Gaffney DK. Pathobiologic characteristics of hereditary breast cancer. *Hum Pathol* 1998;**29**:1140-1144.

132 Malone KE, Daling JR, Ostrander EA. BRCA1 and medullary breast cancer - Reply. *JAMA* 1998;**280**:1227-1228.

133 Eisinger F, Jacquemier J, Charpin C, et al. Mutations at BRCA1: The medullary breast carcinoma revisited. *Cancer Res* 1998;**58**:1588-1592.

134 Agnarsson BA, Jonasson JG, Bjornsdottir IB, Barkardottir RB, Egilsson V, Sigurdsson H. Inherited BRCA2 mutation associated with high grade breast cancer. *Breast Cancer Res Treat* 1998;**47**:121-127.

135 Lakhani SR, Jacquemier J, Sloane JP, et al. Multifactorial analysis of differences between sporadic breast cancers and cancers involving BRCA1 and BRCA2 mutations. *J Natl Cancer Inst* 1998;**90**:1138-1145.

136 Kerlikowske K, Barclay J, Grady D, Sickles EA, Ernster V. Comparison of risk factors for ductal carcinoma in situ and invasive breast cancer. *J Natl Cancer Inst* 1997;**89**:76-82.

137 Sun CC, Lenoir G, Lynch H, Narod SA. In-situ breast cancer and BRCA1. *Lancet* 1996;**348**:408

138 Jacquemier J, Eisinger F, Guinebretiere JM, Stoppa-Lyonnet D, Sobol H. Intraductal component and BRCA1-associated breast cancer. *Lancet* 1996;**348**:1098

139 Malone KE, Daling JR, Weiss NS, Mcknight B, White E, Voigt LF. Family history and survival of young women with invasive breast carcinoma. *Cancer* 1996;**78**:1417-1425.

140 Schouten LJ, Hupperets PSGJ, Jager JJ, et al. Prognostic significance of etiological risk factors in early breast cancer. *Breast Cancer Res Treat* 1997;**43**:217-223.

141 Marcus JN, Watson P, Page DL, et al. Hereditary breast cancer: Pathobiology, prognosis, and BRCA1 and BRCA2 gene linkage. *Cancer* 1996;**77**:697-709.

142 Sigurdsson H, Agnarsson BA, Jonasson JG, et al. Worse survival among breast cancer patients in families carrying BRCA2 susceptibility gene. *Breast Cancer Res Treat* 1996;**37 (Suppl)**:33

143 Robson M, Gilewski T, Haas B, et al. BRCA-associated breast cancer in young women. *J Clin Oncol* 1998;**16**:1642-1649.

144 Verhoog LC, Brekelmans CTM, Seynaeve C, et al. Survival and tumour characteristics of breast-cancer patients with germline mutations of BRCA1. *Lancet* 1998;**351**:316-321.

145 Lee JS, Wacholder S, Struewing JP, et al. Survival after breast cancer in ashkenazi Jewish BRCA1 and BRCA2 mutation carriers. *J Natl Cancer Inst* 1999;**91**:259-263.

146 Ansquer Y, Gautier C, Fourquet A, Asselain B, Stoppa-Lyonnet D. Survival in early-onset BRCA1 breast-cancer patients. *Lancet* 1998;**352**:541

147 Johannsson OT, Ranstam J, Borg A, Olsson H. Survival of BRCA1 breast and ovarian cancer patients: A population- based study from southern Sweden. *J Clin Oncol* 1998;**16**:397-404.

148 Gaffney DK, Brohet RM, Lewis CM, et al. Response to radiation therapy and prognosis in breast cancer patients with BRCA1 and BRCA2 mutations. *Radiother Oncol* 1998;**47**:129-136.

149 Mohammed SN, Smith P, Hodgson SV, et al. Family history and survival in premenopausal breast cancer. *Br J Cancer* 1998;**77**:2252-2256.

150 Harrold EV, Turner BC, Matloff ET, et al. Local recurrence in the conservatively treated breast cancer patient: A correlation with age and family history. *Cancer J Sci Am* 1998;**4**:302-307.

151 Chabner E, Nixon A, Gelman R, et al. Family history and treatment outcome in young women after breast-conserving surgery and radiation therapy for early-stage breast cancer. *J Clin Oncol* 1998;**16**:2045-2051.

152 Haas JA, Schultz DJ, Peterson ME, Solin LJ. An analysis of age and family history on outcome after breast-conservation treatment: The University of Pennsylvania experience. *Cancer J Sci Am* 1998;**4**:308-315.

153 Kerlikowske K, Grady D, Barclay J, Sickles EA, Ernster V. Effect of age, breast density, and family history on the sensitivity of first screening mammography. *JAMA* 1996;**276**:33-38.

154 Law J. Cancers detected and induced in mammographic screening: New screening schedules and younger women with family history. *Br J Radiol* 1997;**70**:62-69.

155 Cox R. Human cancer predisposition and the implications for radiological protection. *Int J Radiat Biol* 1994;**66**:643-647.

156 Adler DD, Wahl RL. New methods for imaging the breast: Techniques, findings, and potential. *Am J Roentgenol* 1995;**164**:19-30.

157 Braeuning MP, Pisano ED. New modalities in breast imaging: Digital mammography and magnetic resonance imaging. *Breast Cancer Res Treat* 1995;**35**:31-38.

158 Howell A, Boggis C, Evans DGR, Lalloo F. Results of mammographic screening for woman with an increased lifetime risk of breast cancer. *Am J Hum Genet* 1996;**59**:A335

159 Rutgers EJT, Hijdendaal MM, Peterse JL, Besnard APE. The yield of an intensive follow-up program for women at increased risk for breast cancer. *Eur J Cancer* 1996;**32A**:27

160 Moller P, Maehle L, Dorum A, et al. Early diagnosis of inherited breast cancer. *Eur J Cancer* 1996;**32A**:28

161 Kollias J, Sibbering DM, Holland PAM, et al. Results of screening for women aged <50 with a family hostory of breast cancer. *Eur J Surg Oncol* 1996;**22**:551

162 Lalloo F, Boggis C, Evans DGR, Howell A. Screening for breast cancer in women with a family history. *Eur J Hum Genet* 1996;**4**:43

163 McLeish L, Goudie D, Preece P, Reis M, Steel M, Faed M. Breast cancer detection among women followed up or assessed because of their family history. *Eur J Hum Genet* 1996;**4**:45

164 Heap E, Bradburn M, Fentiman I, Hodgson S. A review of the effect of surveillance on outcome for women considered to be at increased risk of developing breast cancer. *J Med Genet* 1996;**33**:S39

165 Tilanus-Linthorst MMA, Bartels CCM, Obdeijn AIM, Kuenen-Bouwmeester V, Klijn JGM, Oudkerk M. Gunstige resultaten van periodieke controle bij vrouwen met verhoogd rsisico van borstkanker. *Ned Tijdschr Geneeskd* 1995;**139**:445-449.

166 Houlston RS, Lemoine L, McCarter E, et al. Screening and genetic counselling for relatives of patients with breast cancer in a family cancer clinic. *J Med Genet* 1992;**29**:691-694.

167 Moller P, Maehle L, Heimdal K, et al. Inherited breast carcinoma - Prospective findings in 1 194 women at risk. *Acta Oncol* 1996;**35**:7-11.

168 Chart PL, Franssen E. Management of women at increased risk for breast cancer: preliminary results from a new program. *Can Med Assoc J* 1997;**157**:1235-1242.

169 Lalloo F, Boggis CRM, Evans DGR, Shenton A, Threlfall AG, Howell A. Screening by mammography, women with a family history of breast cancer. *Eur J Cancer* 1998;**34**:937-940.

170 Kollias J, Sibbering DM, Blamey RW, et al. Screening women aged less than 50 years with a family history of breast cancer. *Eur J Cancer* 1998;**34**:878-883.

171 Vasen HFA. Screening in breast cancer families: is it useful? *Ann Med* 1994;**26**:185-190.

172 Burke W, Daly M, Garber J, et al. Recommendations for follow-up care of individuals with an inherited predisposition to cancer: 2. BRCA1 and BRCA2. *JAMA* 1997;**277**:997-1003.

173 Eisinger F, Alby N, Bremond A, et al. Recommendations for medical management of hereditary breast and ovarian cancer: the french national ad hoc committee. *Ann Oncol* 1998;**9**:939-950.

174 Vasen HFA, Haites NE, Evans DGR, et al. Current policies for surveillance and management in women at risk of breast and ovarian cancer: a survey among 16 european family cancer clinics. *Eur J Cancer* 1998;**34**:1922-1926.

175 Stefanek ME. Bilateral prophylactic mastectomy: issues and concerns. *J Natl Cancer Inst Mono* 1995;**17**:37-42.

176 Bilimoria MM, Morrow M. The woman at increased risk for breast cancer: evaluation and management strategies. *CA Cancer J Clin* 1995;**45**:263-278.

177 Lopez MJ, Porter KA. The current role of prophylactic mastectomy. *Surg Clin North Am* 1996;**76**:231

178 Schrag D, Kuntz KM, Garber JE, Weeks JC. Decision analysis-effects of prophylactic mastectomy and oophorectomy on life expectancy among women with BRCA1 or BRCA2 mutations. *N Engl J Med* 1997;**336**:1465-1471.

179 Grann VR, Panageas KS, Whang W, Antman KH, Neugut AI. Decision analysis of prophylactic mastectomy and oophorectomy in BRCA1-positive or BRCA2-positive patients. *J Clin Oncol* 1998;**16**:979-985.

180 Tengs TO, Winer EP, Paddock S, Aguilar-Chavez O, Berry DA. Testing for the BRCA1 and BRCA2 breast-ovarian cancer susceptibility genes. *Med Decision Making* 1998;**18**:365-375.

181 Hartmann LC, Schaid DJ, Woods JE, et al. Efficacy of bilateral prophylactic mastectomy in women with a family history of breast cancer. *N Engl J Med* 1999;**340**:77-84.

182 Fisher B, Costantino JP, Wickerham DL, et al. Tamoxifen for prevention of breast cancer: Report of the National Surgical Adjuvant Breast and Bowel Project P-1 study. *J Natl Cancer Inst* 1998;**90**:1371-1388.

183 Osin P, Crook T, Powles T, Peto J, Gusterson B. Hormone status of in-situ cancer in BRCA1 and BRCA2 mutation carriers. *Lancet* 1998;**351**:1487

184 Osin P, Gusterson BA, Philp E, et al. Predicted anti-oestrogen resistance in BRCA-associated familial breast cancers. *Eur J Cancer* 1998;**34**:1683-1686.

185 Wagner TMU, Moslinger RA, Muhr D, et al. BRCA1-related breast cancer in Austrian breast and ovarian cancer families: Specific BRCA1 mutations and pathological characteristics. *Int J Cancer* 1998;**77**:354-360.

186 Loman N, Johannsson O, Bendahl PO, Borg A, Ferno M, Olsson H. Steroid receptors in hereditary breast carcinomas associated with BRCA1 or BRCA2 mutations or unknown susceptibility genes. *Cancer* 1998;**83**:310-319.

187 Furgyik S, Grubb R, Kullander S, Sandahl B, Wingerup L, Eydal A. Familial occurrence of cervical cancer, stages 0-IV. *Acta Obstet Gynecol Scand* 1986;**65**:223-227.

188 Brinton LA, Tashima KT, Lehman HF, et al. Epidemiology of cervical cancer by cell type. *Cancer Res* 1987;**47**:1706-1711.

189 Bruinse HW, te Velde ER, de Gast BC. Human leukocyte antigen patterns in a family with cervical cancer. *Gynecol Oncol* 1981;**12**:249-252.

190 Bender S. Carcinoma in-situ of cervix in sisters. *Br Med J* 1976;**i**:502

191 Ahlbom A, Lichtenstein P, Malmstrom H, Feychting M, Hemminki K, Pedersen NL. Cancer in twins: Genetic and nongenetic familial risk factors. *J Natl Cancer Inst* 1997;**89**:287-293.

192 Cannon L, Bishop DT, Skolnick M, Hunt S, Lyon JL, Smart CR. Genetic epidemiology of prostate cancer in the Utah Mormon genealogy. *Cancer Surv* 1982;**1**:47-69.

193 Kessler II, Kulcar Z, Zimolo A, Grgurevic M, Strnad M, Goodwin BJ. Cervical cancer in Yugoslavia. II. Epidemiologic factors of possible etiologic significance. *J Natl Cancer Inst* 1974;**53**:51-60.

194 Auranen A, Grenman S, Makinen J, Pukkala E, Sankila R, Salmi T. Borderline ovarian tumors in Finland: Epidemiology and familial occurrence. *Am J Epidemiol* 1996;**144**:548-553.

195 Giardiello FM, Welsh SB, Hamilton SR, et al. Increased risk of cancer in the Peutz-Jeghers syndrome. *N Engl J Med* 1987;**24**:1511-1514.

196 Yamakazi S, Katayama I, Yokozeki H, Nishioka K. Letter to the editor: Reproductive tract tumours in Peutz-Jeghers syndrome. *Br J Dermatol* 1993;**128**:466-467.

197 Clement PB, Young RH, Scully RE. Clinical syndromes associated with tumors of the female genital tract. *Semin Diagn Pathol* 1991;**8**:204-233.

198 Podczaski E, Kaminski PF, Pees RC, Singapuri K, Sorosky JI. Case report: Peutz-Jeghers syndrome with ovarian sex cord tumor with annular tubules and cervical adenoma malignum. *Gynecol Oncol* 1991;**42**:74-78.

199 Gilks CB, Young RH, Aguirre P, Delellis RA, Scully RE. Adenoma malignum (minimal deviation adenocarcinoma) of the cervix: a clinicopathological and immunohistochemical analysis of 26 cases. *Am J Surg Pathol* 1989;**13**:717-729.

200 Hodgson SV, Maher ER. *A practical guide to human cancer genetics*. Cambridge: Cambridge University Press, 1993;

201 German J. Bloom's syndrome .XX. The first 100 cancers. *Cancer Genet Cytogenet* 1997;**93**:100-106.

202 Alter BP. Fanconi's anemia and malignancies. *Am J Hematol* 1996;**53**:99-110.

203 Drachtman RA, Alter BP. Dyskeratosis congenita: clinical and genetic heterogeneity: report of a new case and review of the literature. *Am J Ped Hematol Oncol* 1992;**14**:297-304.

204 Hizawa K, Iida M, Matsumoto T, et al. Cancer in Peutz-Jeghers syndrome. *Cancer* 1993;**72**:2777-2781.

205 Spiegelman AD, Arese P, Phillips RKS. Polyposis: the Peutz-Jeghers syndrome. *Br J Surg* 1995;**82**:1311-1314.

206 Wank R, Thomssen C. High risk of squamous cell carcinoma of the cervix for women with HLA-DQw3. *Nature* 1991;**352**:723-725.

207 Odunsi K, Terry G, Ho L, Bell J, Cuzick J, Ganesan TS. Association between HLA DQB1 03 and cervical intra- epithelial neoplasia. *Mol Med* 1995;**1**:161-171.

208 Apple RJ, Erlich HA, Klitz W, Manos MM, Becker TM, Wheeler CM. HLA DR-DQ associations with cervical carcinoma show papillomavirus-type specificity. *Nat Genet* 1994;**6**:157-162.

209 Sanjeevi CB, Hjelmstrom P, Hallmans G, et al. Different HLA-DR-DQ haplotypes are associated with cervical intraepithelial neoplasia among human papillomavirus type-16 seropositive and ronegative Swedish women. *Int J Cancer* 1996;**68**:409-414.

210 Odunsi K, Terry G, Ho L, Bell J, Cuzick J, Ganesan TS. Susceptibility to human papillomavirus-associated cervical intra-epithelial neoplasia is determined by specific HLA DR-DQ alleles. *Int J Cancer* 1996;**67**:595-602.

211 Sastre-Garau X, Loste MN, Vincent-Salomon A, et al. Decreased frequency of HLA-DRB1*13 alleles in Frenchwomen with HPV- positive carcinoma of the cervix. *Int J Cancer* 1996;**69**:159-164.

212 Allen M, Kalantari M, Ylitalo N, et al. HLA DQ-DR haplotype and susceptibility to cervical carcinoma: Indications of increased risk for development of cervical carcinoma in individuals infected with HPV 18. *Tissue Antigens* 1996;**48**:32-37.

213 Lovett E. Family studies in cancer of the colon and rectum. *Br J Surg* 1976;**63**:13-18.

214 Stephenson BM, Finan PJ, Gascoyne J, Garbett F, Murday VA, Bishop DT. Frequency of familial colorectal cancer. *Br J Surg* 1991;**78**:1162-1166.

215 Kotake K, Koyama Y, Nasu J, Fukutomi T, Yamaguchi N. Relation of family history of cancer and environmental factors to the risk of colorectal cancer: a case-control study. *Jpn J Clin Oncol* 1995;**25**:195-202.

216 Fuchs CS, Giovannucci EL, Colditz GA, Hunter DJ, Speizer FE, Willett WC. A prospective study of family history and the risk of colorectal cancer. *N Engl J Med* 1994;**331**:1669-1674.

217 Burt RW. Familial risk and colon cancer. *Int J Cancer* 1996;**69**:44-46.

218 Hall NR, Finan PJ, Ward B, Turner G, Bishop DT. Genetic susceptibility to colorectal cancer in patients under 45 years of age. *Br J Surg* 1994;**81**:1485-1489.

219 Slattery ML, Kerber RA. Family history of cancer and colon cancer risk: The Utah population database. *J Natl Cancer Inst* 1994;**86**:1618-1626.

220 St.John DJB, McDermott FT, Hopper JL, Debney EA, Johnson WR, Hughes ES. Cancer risk in relatives of patients with common colorectal cancer. *Ann Intern Med* 1993;**118**:785-790.

221 Carstensen B, Soll-Johanning H, Villadsen E, Sondergaard JO, Lynge E. Familial aggregation of colorectal cancer in the general population. *Int J Cancer* 1996;**68**:428-435.

222 Papp J, Csokay B, Bosze P, et al. Allele loss from large regions of chromosome 17 is common only in certain histological subtypes of ovarian carcinomas. *Br J Cancer* 1996;**74**:1592-1597.

223 Rozen P, Fireman Z, Figer A, Legum C, Ron E, Lynch HT. Family history of colorectal cancer as a marker of potential malignancy within a screening program. *Cancer* 1987;**60**:248-254.

224 Duncan JL, Kyle J. Family incidence of carcinoma of the colon and rectum in north-east Scotland. *Gut* 1982;**23**:169-171.

225 Kune GA, Kune S, Watson LF. Children, age at first birth, and colorectal cancer risk: data from the Melbourne colorectal cancer study. *Am J Epidemiol* 1989;**129**:533-542.

226 Cannon-Albright LA, Skolnick MH, Bishop DT, Lee RG, Burt RW. Common inheritance of susceptibility to colonic adenomatous polyps and associated colorectal cancers. *N Engl J Med* 1988;**319**:533-537.

227 Bonelli L, Martines H, Conio M, Bruzzi P, Aste H. Family history of colorectal cancer as a risk factor for menign and malignant tumours of the large bowel. a case-control study. *Int J Cancer* 1988;**41**:513-517.

228 Pedersen KM. Economics of cancer screening: Total costs and benefits in economic terms. *Eur J Cancer* 1994;**30A**:879-884.

229 Compton JG, Goldstein AM, Turner M, et al. Fine mapping of the locus for nevoid basal cell carcinoma syndrome on chromosome 9q. *J Invest Dermatol* 1994;**103**:178-181.

230 Winawer SJ, Zauber AG, Gerdes H, et al. Risk of colorectal cancer in the families of patients with adenomatous polyps. *N Engl J Med* 1996;**334**:82-87.

231 Aitken JF, Bain CJ, Ward M, Siskind V, Maclennan R. Risk of colorectal adenomas in patients with a family history of colorectal cancer: Some implications for screening programmes. *Gut* 1996;**39**:105-108.

232 Boutron MC, Faivre J, Quipourt V, Senesse P, Michiels C. Family history of colorectal tumours and implications for the adenoma-carcinoma sequence: a case control study. *Gut* 1995;**37**:830-834.

233 Bazzoli F, Fossi S, Sottili S, et al. The risk of adenomatous polyps in asymptomatic first-degree relatives of persons with colon cancer. *Gastroenterology* 1995;**109**:783-788.

234 Ahsan H, Neugut AI, Garbowski GC, et al. Family history of colorectal adenomatous polyps and increased risk for colorectal cancer. *Ann Intern Med* 1998;**128**:900

235 Pariente A, Milan C, Lafon J, Faivre J. Colonoscopic screening in first-degree relatives of patients with 'sporadic' colorectal cancer: A case-control study. *Gastroenterology* 1998;**115**:7-12.

236 Kerber RA, Slattery ML, Potter JD, Caan BJ, Edwards SL. Risk of colon cancer associated with a family history of cancer or colorectal polyps: The diet, activity, and reproduction in colon cancer study. *Int J Cancer* 1998;**78**:157-160.

237 Fernandez E, La Vecchia C, D'Avanzo B, Negri E, Franceschi S. Risk factors for colorectal cancer in subjects with family history of the disease. *Br J Cancer* 1997;**75**:1381-1384.

238 Le Marchand L, Wilkens LR, Hankin JH, Kolonel LN, Lyu LC. Independent and joint effects of family history and lifestyle on colorectal cancer risk: implications for prevention. *Cancer Epidemiol Biomarkers Prev* 1999;**8**:45-51.

239 Houlston RS, Murday V, Harocopos C, Williams CB, Slack J. Screening and genetic counselling for relatives of patients with colorectal cancer in a family cancer clinic. *Br Med J* 1990;**301**:366-368.

240 Houlston R, Path MRC, Bourne TH, et al. Use of family history in a screening clinic for familial ovarian cancer. *Gynecol Oncol* 1992;**47**:247-252.

241 Nelson CL, Sellers TA, Rich SS, Potter JD, Mcgovern PG, Kushi LH. Familial clustering of colon, breast, uterine, and ovarian cancers as assessed by family history. *Genet Epidemiol* 1993;**10**:235-244.

242 Damber L, Gronberg H, Damber JE. Familial prostate cancer and possible associated malignancies: Nation- wide register cohort study in Sweden. *Int J Cancer* 1998;**78**:293-297.

243 Rustgi AK. Medical Progress: Hereditary gastrointestinal polyposis and nonpolyposis syndromes. *N Engl J Med* 1994;**331**:1694-1702.

244 Murday V, Slack J. Inherited disorders associated with colorectal cancer. *Cancer Surv* 1989;**1**:139-157.

245 Finan MC, Ray MK. Gastrointestinal polyposis syndromes. *Dermatol Clin* 1989;**7**:419-434.

246 Luk GD. Diagnosis and therapy of hereditary polyposis syndromes. *Gastroenterologist* 1995;**3**:153-167.

247 Watson P, Lynch HT. The tumor spectrum in HNPCC. *Anticancer Res* 1994;**14**:1635-1639.

248 Mecklin JP, Jarvinen HJ. Tumor spectrum in Cancer Family Syndrome (Hereditary Nonpolyposis Colorectal Cancer). *Cancer* 1991;**68**:1109-1112.

249 Dewaard F, Mouridsen H. Breast cancer screening: Methods, human benefits, human costs. *Eur J Cancer* 1994;**30A**:875-877.

250 Burn J, Chapman PD, Eastham EJ. Familial adenomatous polyposis. *Arch Dis Child* 1994;**71**:103-105.

251 Vasen HFA, Wijnen JT, Menko FH, et al. Cancer risk in families with hereditary nonpolyposis colorectal cancer diagnosed by mutation analysis. *Gastroenterology* 1996;**110**:1020-1027.

252 Lynch HT, Smyrk T. Hereditary nonpolyposis colorectal cancer (Lynch syndrome) - An updated review. *Cancer* 1996;**78**:1149-1167.

253 Marra G, Boland CR. Hereditary nonpolyposis colorectal cancer: the syndrome, the genes, and historical perspectives. *J Natl Cancer Inst* 1995;**87**:1114-1125.

254 Dunlop MG, Farrington SM, Carothers AD, et al. Cancer risk associated with germline DNA mismatch repair gene mutations. *Hum Mol Genet* 1997;**6**:105-110.

255 Fishel R, Lescoe MK, Rao MRS, et al. The human mutator gene homolog MSH2 and its association with hereditary nonpolyposis colon cancer. *Cell* 1993;**75**:1027-1038.

256 Nicolaides NC, Papadopoulos N, Liu B, et al. Mutations of two PMS homologues in hereditary nonpolyposis colon cancer. *Nature* 1994;**371**:75-80.

257 Leach FS, Nicolaides NC, Papadopoulos N, et al. Mutations of a mutS homolog in hereditary nonpolyposis colorectal cancer. *Cell* 1993;**75**:1215-1225.

258 Papadopoulos N, Nicolaides NC, Wei YF, et al. Mutation of a mutL homolog in hereditary colon cancer. *Science* 1994;**263**:1625-1629.

259 Bronner CE, Baker SM, Morrison PT, et al. Mutation in the DNA mismatch repair gene homologue hMLH1 is associated with hereditary non-polyposis colon cancer. *Nature* 1994;**368**:258-261.

260 Akiyama Y, Sato H, Yamada T, et al. Germ-line mutation of the hMSH6/GTBP gene in an atypical hereditary nonpolyposis colorectal cancer kindred. *Cancer Res* 1997;**57**:3920-3923.

261 Liu B, Parsons R, Papadopoulos N, et al. Analysis of mismatch repair genes in hereditary non-polyposis colorectal cancer patients. *Nat Med* 1996;**2**:169-174.

262 Wijnen JT, Vasen HFA, Meera Khan P, et al. Clinical findings with implications for genetic testing in families with clustering of colorectal cancer. *N Engl J Med* 1998;**339**:511-518.

263 Rhyu MS. Molecular mechanisms underlying hereditary nonpolyposis colorectal carcinoma. *J Natl Cancer Inst* 1996;**88**:240-251.

264 Umar A, Kunkel TA. DNA-replication fidelity, mismatch repair and genome instability in cancer cells. *Eur J Biochem* 1996;**238**:297-307.

265 Parsons R, Myeroff LL, Liu B, et al. Microsatellite instability and mutations of the transforming growth factor beta type II receptor gene in colorectal cancer. *Cancer Res* 1995;**55**:5548-5550.

266 Eshleman JR, Markowitz SD. Mismatch repair defects in human carcinogenesis. *Hum Mol Genet* 1996;**5**:1489-1494.

267 Mellon I, Rajpal DK, Koi M, Boland CR, Champe GN. Transcription-coupled repair deficiency and mutations in human mismatch repair genes. *Science* 1996;**272**:557-560.

268 Holzman D. Mismatch repair genes matched to several new roles in cancer. *J Natl Cancer Inst* 1996;**88**:950-951.

269 Parsons R, Li GM, Longley M, et al. Mismatch repair deficiency in phenotypically normal human cells. *Science* 1995;**268**:738-740.

270 Aaltonen LA, Peltomaki P, Mecklin JP, et al. Replication errors in benign and malignant tumors from hereditary nonpolyposis celorectal cancer patients. *Cancer Res* 1994;**54**:1645-1648.

271 Eshleman JR, Markowitz SD. Microsatellite instability in inherited and sporadic neoplasms. *Curr Opin Oncol* 1995;**7**:83-89.

272 Aaltonen LA, Salovaara R, Kristo P, et al. Incidence of hereditary nonpolyposis colorectal cancer and the feasibility of molecular screening for the disease. *N Engl J Med* 1998;**338**:1481-1487.

273 Jass JR, Cottier DS, Jeevaratnam P, et al. Diagnostic use of microsatellite instability in hereditary non-polyposis colorectal cancer. *Lancet* 1995;**346**:1200-1201.

274 Moslein G, Tester DJ, Lindor NM, et al. Microsatellite instability and mutation analysis of hMSH2 and hMLH1 in patients with sporadic, familial and hereditary colorectal cancer. *Hum Mol Genet* 1996;**5**:1245-1252.

275 Baba S. Clinical applications of genetic studies in hereditary colorectal cancer. In: Muller H, Scott RJ, Weber W, eds. *Hereditary Cancer*. Basel: Karger, 1996;173-190.

276 Beck NE, Tomlinson IP, Homfray T, Hodgson SV, Harocopos CJ, Bodmer WF. Genetic testing is important in families with a history suggestive of hereditary non-polyposis colorectal cancer even if the Amsterdam criteria are not fulfilled. *Br J Surg* 1997;**84**:233-237.

277 Liu B, Farrington SM, Petersen GM, et al. Genetic instability occurs in the majority of young patients with colorectal cancer. *Nat Med* 1995;**1**:348-352.

278 Rodriguez-Bigas MA, Boland CR, Hamilton SR, et al. A National Cancer Institute workshop on hereditary nonpolyposis colorectal cancer syndrome: Meeting highlights and Bethesda guidelines. *J Natl Cancer Inst* 1997;**89**:1758-1762.

279 Lynch HT, Smyrk TC. Identifying hereditary nonpolyposis colorectal cancer. *N Engl J Med* 1998;**338**:1537-1538.

280 Lynch HT, Smyrk T. Colorectal cancer, survival advantage, and hereditary nonpolyposis colorectal carcinoma. *Gastroenterology* 1996;**110**:943-947.

281 Sankila R, Aaltonen LA, Jarvinen HJ, Mecklin JP. Better survival rates in patients with MLH1-associated hereditary colorectal cancer. *Gastroenterology* 1996;**110**:682-687.

282 Aarnio M, Mustonen H, Mecklin JP, Jarvinen HJ. Prognosis of colorectal cancer varies in different high-risk conditions. *Ann Med* 1998;**30**:75-80.

283 Watson P, Lin KM, Rodriguez-Bigas MA, et al. Colorectal carcinoma survival among hereditary nonpolyposis colorectal carcinoma family members. *Cancer* 1998;**83**:259-266.

284 Percesepe A, Benatti P, Roncucci L, et al. Survival analysis in families affected by hereditary non-polyposis colorectal cancer. *Int J Cancer* 1997;**71**:373-376.

285 Bertario L, Russo A, Sala P, et al. Survival of patients with hereditary colorectal cancer: Comparison of HNPCC and colorectal cancer in FAP patients with sporadic colorectal cancer. *Int J Cancer* 1999;**80**:183-187.

286 Jass JR, Pokos V, Arnold JL, et al. Colorectal neoplasms detected colonoscopically in at-risk members of colorectal cancer families stratified by the demonstration of DNA microsatellite instability. *J Mol Med* 1996;**74**:547-551.

287 Muta H, Noguchi M, Perucho M, et al. Clinical implications of microsatellite instability in colorectal cancers. *Cancer* 1996;**77**:265-270.

288 Samowitz WS, Slattery ML, Kerber RA. Microsatellite instability in human colonic cancer is not a useful clinical indicator of familial colorectal cancer. *Gastroenterology* 1995;**109**:1765-1771.

289 Peltomaki P. Microsatellite instability as an indicator of hereditary susceptibility to colon cancer. *Gastroenterology* 1995;**109**:2031-2033.

290 Hunt LM, Rooney PS, Bostock K, Robinson MHE, Hardcastle JD, Armitage NC. Chemical and immunological testing for faecal occult blood in screening subjects at risk of familial colorectal cancer. *Gut* 1997;**40**:110-112.

291 Gaglia P, Atkin WS, Whitelaw S, et al. Variables associated with the risk of colorectal adenomas in asymptomatic patients with a family history of colorectal cancer. *Gut* 1995;**36**:385-390.

292 Guillem JG, Neugut AI, Forde KA, Waye JD, Treat MR. Colonic neoplasms in asymptomatic firts-degree relatives of colon cancer patients. *Am J Gastroenterol* 1988;**83**:271-273.

293 Wu CS, Tung SY, Chen PC, Kuo YC. The role of colonoscopy in screening persons with family history of colorectal cancer. *J Gastroenterol Hepatol* 1995;**10**:319-323.

294 Vasen HFA, Griffioen G, Offerhaus GJA, et al. The value of screening and central registration of families with familial adenomatous polyposis. *Dis Colon Rectum* 1990;**33**:227-230.

295 Church J. Hereditary colon cancers can be tiny: a cautionary case report of the results of colonoscopic surveillance. *Am J Gastroenterol* 1998;**93**:2289-2290.

296 Ahlquist DA. Aggressive polyps in hereditary nonpolyposis colorectal cancer: Targets for screening. *Gastroenterology* 1995;**108**:1590-1592.

297 Jarvinen HJ, Mecklin JP, Sistonen P. Screening reduces colorectal cancer rate in families with hereditary nonpolyposis colorectal cancer. *Gastroenterology* 1995;**108**:1405-1411.

298 Syngal SS, Weeks JC, Schrag D, Garber JE, Kuntz KM. Benefits of colonoscopic surveillance and prophylactic colectomy in patients with hereditary nonpolyposis colorectal cancer mutations. *Ann Intern Med* 1998;**129**:787-796.

299 Alberts DS, Lipkin M, Levin B. Genetic screening for colorectal cancer and intervention. *Int J Cancer* 1996;**69**:62-63.

300 Blackburn GL, Giardiello FM. Developing strategies for intervention/prevention trials of individuals at risk of hereditary colon cancer. *J Natl Cancer Inst Mono* 1995;107-110.

301 Marcus AJ. Aspirin as prophylaxis against colorectal cancer [editorial; comment]. *N Engl J Med* 1995;**333**:656-658.

302 Berkel HJ, Holcombe RF, Middlebrooks M, Kannan K. Nonsteroidal antiinflammatory drugs and colorectal cancer. *Epidemiol Rev* 1996;**18**:205-217.

303 Decosse JJ, Miller HH, Lesser ML. Effect of wheat fiber and vitamins C and E on rectal polyps in patients with familial adenomatous polyposis. *J Natl Cancer Inst* 1989;**81**:1290-1297.

304 Giardiello FM, Hamilton SR, Krush AJ, et al. Treatment of colonic and rectal adenomas with sulindac in familial adenomatous polyposis. *N Engl J Med* 1993;**328**:1313-1316.

305 Burn J, Chapman PD, Mathers J, et al. The protocol for a European double-blind trial of aspirin and resistant starch in familial adenomatous polyposis: The CAPP Study. *Eur J Cancer* 1995;**31A**:1385-1386.

306 Burn J, Alonso A, Bertario L, et al. Concerted action polyp prevention, CAPP2: A trial of aspirin and/or resistant starch in people at risk of hereditary non-polyposis colon cancer (HNPCC). In: Baig SS, ed. *Cancer Research Supported Under Biomed I.* 1013 CN Amsterdam: I O S Press, 1998;17-23.

307 Cats A, Kleibeuker JH, van der Meer R, et al. Randomized, double-blinded, placebo-controlled intervention study with supplemental calcium in families with hereditary nonpolyposis colorectal cancer. *J Natl Cancer Inst* 1995;**87**:598-603.

308 Stern HS, Gregoire RC, Kashtan H, Stadler J, Bruce RW. Long-term effects of dietary calcium on risk markers for colon cancer in patients with familial polyposis. *Surgery* 1990;**108**:528-533.

309 Giardiello FM, Offerhaus JA, Tersmette AC, et al. Sulindac induced regression of colorectal adenomas in familial adenomatous polyposis: Evaluation of predictive factors. *Gut* 1996;**38**:578-581.

310 Stiff GJ, Alwafi A, Jenkins H, Lari J. Management of infantile polyposis syndrome. *Arch Dis Child* 1995;**73**:253-254.

311 Nugent KP, Northover J. Total colectomy and ileorectal anastomosis. In: Phillips RKS, Spigelman AD, Thomson JPS, eds. *Familial Adenomatous Polyposis and other polyposis syndromes.* London: Edward Arnold, 1994;79-91.

312 Phillips RKS, Spigelman AD. Can we safely delay or avoid prophylactic colectomy in familial adenomatous polyposis? *Br J Surg* 1996;**83**:769-770.

313 Setti-Carraro P, Nicholls RJ. Choice of prophylactic surgery for the large bowel component of familial adenomatous polyposis. *Br J Surg* 1996;**83**:885-892.

314 Lynch HT. Is there a role for prophylactic subtotal colectomy among hereditary nonpolyposis colorectal cancer germline mutation carriers? *Dis Colon Rectum* 1996;**39**:109-110.

315 Rodriguez-Bigas MA. Prophylactic colectomy for gene carriers in hereditary nonpolyposis colorectal cancer: Has the time come? *Cancer* 1996;**78**:199-201.

316 Church JM. Prophylactic colectomy in patients with hereditary nonpolyposis colorectal cancer. *Ann Med* 1996;**28**:479-482.

317 Desai DC, Neale KF, Talbot IC, Hodgson SV, Phillips RKS. Juvenile polyposis. *Br J Surg* 1995;**82**:14-17.

318 Blum HE, Wieland S, von Weizsacker F. Gene therapy: Basic concepts and applications in gastrointestinal diseases. *Digestion* 1997;**58**:87-97.

319 Westbrook CA, Arenas RB. Gene therapy of the gut: Introduction of the APC tumor-suppressor gene for cancer prevention or treatment. *Advan Drug Delivery Rev* 1995;**17**:349-355.

320 Parazzini F, Lavecchia C, Moroni S, Chatenoud L, Ricci E. Family history and the risk of endometrial cancer. *Int J Cancer* 1994;**59**:460-462.

321 Suomi R, Hakalaalapietila T, Leminen A, Mecklin JP, Lehtovirta P. Hereditary aspects of endometrial adenocarcinoma. *Int J Cancer* 1995;**62**:132-137.

322 Gruber SB, Thomson WD, the Cancer and Steroid Hormone Study Group. A population-based study of endometrial cancer and familial risk in younger women. *Cancer Epidemiol Biomarkers Prev* 1996;**5**:411-417.

323 Slattery ML, Mori M, Gao R, Kerber RA. Impact of family history of colon cancer on development of multiple primaries after diagnosis of colon cancer. *Dis Colon Rectum* 1995;**38**:1053-1058.

324 Schildkraut JM, Risch N, Thompson WD. Evaluating genetic association among ovarian, breast, and endometrial cancer: evidence for a breast/ovarian cancer relationship. *Am J Hum Genet* 1989;**45**:521-529.

325 Peto J, Easton DF, Matthews FE, Ford D, Swerdlow AJ. Cancer mortality in relatives of women with breast cancer: the OPCS Study. Office of Population Censuses and Surveys. *Int J Cancer* 1996;**65**:275-283.

326 Sandles LG, Shulman LP, Elias S, et al. Endometrial adenocarcinoma: genetic analysis suggesting heritable site-specific uterine cancer. *Gynecol Oncol* 1992;**47**:167-171.

327 Boltenberg A, Furgyik S, Kullander S. Familial cancer aggregation in cases of adenocarcinoma corporis uteri. *Acta Obstet Gynecol Scand* 1990;**69**:249-258.

328 Shulman LP, Photopulos GJ, Simpson JL, Smiley LM, Sandles LG, Elias S. Adenocarcinoma of the endometrium: family cancer studies demonstrating heritable site-specific endometrial cancer. *Am J Hum Genet* 1992;**51**:A50

329 Lynch HT, Lynch J, Conway T, Watson P, Coleman RL. Familial aggregation of carcinoma of the endometrium. *Am J Obstet Gynecol* 1994;**171**:24-27.

330 Menko FH, Verheijen RHM, Everhardt E, et al. Endometrial cancer in four sisters: report of a kindred with presumed cancer family syndrome. *Gynecol Oncol* 1994;**54**:171-174.

331 Watson P, Vasen HFA, Mecklin JP, Jarvinen H, Lynch HT. The risk of endometrial cancer in hereditary nonpolyposis colorectal cancer. *Am J Med* 1994;**96**:516-520.

332 Aarnio M, Mecklin JP, Aaltonen LA, Nystrom-Lahti M, Jarvinen HJ. Life-time risk of different cancers in hereditary non- polyposis colorectal cancer (HNPCC) syndrome. *Int J Cancer* 1995;**64**:430-433.

333 Vasen HFA, Watson P, Mecklin JP, et al. The epidemiology of endometrial cancer in hereditary nonpolyposis colorectal cancer. *Anticancer Res* 1994;**14**:1675-1678.

334 Watson P, Lynch HT. Extracolonic cancer in Hereditary Nonpolyposis Colorectal Cancer. *Cancer* 1993;**71**:677-685.

335 Weber T. Clinical surveillance recommendations adopted for HNPCC. *Lancet* 1996;**348**:465

336 Schlehofer B, Pommer W, Mellemgaard A, et al. International renal-cell-cancer study .6. The role of medical and family history. *Int J Cancer* 1996;**66**:723-726.

337 Neumann HPH, Zbar B. Renal cysts, renal cancer and von Hippel-Lindau disease. *Kidney Int* 1997;**51**:16-26.

338 Maher ER, Yates JRW. Familial renal cell carcinoma: clinical and molecular genetic aspects. *Br J Cancer* 1991;**63**:176-179.

339 Levinson AK, Johnson DE, Strong LC, Pathak S, Huff V, Saunders GF. Familial renal cell carcinoma: hereditary or coincidental? *J Urol* 1990;**144**:849-851.

340 Cohen AJ, Li FP, Berg S, et al. Hereditary renal-cell carcinoma associated with a chromosomal translocation. *N Engl J Med* 1979;**301**:592-595.

341 Kovacs G, Hoene E. Loss of der(3) in renal carcinoma cells of a patient with constitutional t(3;12). *Hum Genet* 1988;**78**:148-150.

342 Kovacs G, Brusa P, de Riese W. Tissue-specific expression of a constitutional 3;6 translocation: development of multiple bilateral renal-cell carcinoma. *Int J Cancer* 1989;**43**:422-427.

343 Li FP, Decker HJH, Zbar B, et al. Clinical and genetic studies of renal cell carcinomas in a family with a constitutional chromosome 3;8 translocation. *Ann Intern Med* 1993;**118**:106-111.

344 Koolen MI, van der Meyden PM, Bodmer D, et al. A familial case of renal cell carcinoma and a t(2;3) chromosome translocation. *Kidney Int* 1998;**53**:273-275.

345 Gemmill RM, West JD, Boldog F, et al. The hereditary renal cell carcinoma 3;8 translocation fuses FHIT to a patched-related gene, TRC8. *Proc Natl Acad Sci U S A* 1998;**95**:9572-9577.

346 Teh BT, Giraud S, Sari NF, et al. Familial non-VHL non-papillary clear-cell renal cancer. *Lancet* 1997;**349**:848-849.

347 Zbar B, Tory K, Merino M, et al. Hereditary papillary renal cell carcinoma. *J Urol* 1994;**151**:561-566.

348 Zbar B, Glenn G, Lubensky I, et al. Hereditary papillary renal cell carcinoma: Clinical studies in 10 families. *J Urol* 1995;**153**:907-912.

349 Zbar B, Glenn G, Lubensky I, et al. Letters to the editor: Hereditary papillary renal cell carcinoma: clinical studies in 10 families. *J Urol* 1996;**156**:1781

350 Schmidt L, Duh FM, Chen F, et al. Germline and somatic mutations in the tyrosine kinase domain of the MET proto-oncogene in papillary renal carcinomas. *Nat Genet* 1997;**16**:68-73.

351 Cook JA, Oliver K, Mueller RF, Sampson J. A cross sectional study of renal involvement in tuberous sclerosis. *J Med Genet* 1996;**33**:480-484.

352 Pritchard-Jones K. Wilms tumour and other genetic causes of renal cancer. In: Eeles RA, Ponder BAJ, Easton DF, Horwich A, eds. *Genetic predisposition to cancer*. London: Chapman & Hall medical, 1996;105-122.

353 Bjornsson J, Short MP, Kwiatkowski DJ, Henske EP. Tuberous sclerosis-associated renal cell carcinoma - Clinical, pathological, and genetic features. *Am J Pathol* 1996;**149**:1201-1208.

354 Lynch HT, Ens JA, Lynch JF. The Lynch syndrome II and urological malignancies. *J Urol* 1990;**143**:24-28.

355 Teh BT, Farnebo F, Kristoffersson U, et al. Autosomal dominant primary hyperparathyroidism and jaw tumor syndrome associated with renal hamartomas and cystic kidney disease: Linkage to 1q21-q32 and loss of the wild type allele in renal hamartomas. *J Clin Endocrinol Metab* 1996;**81**:4204-4211.

356 Greenberg F, Copeland K, Gresik MV. Expanding the spectrum of the Perlman syndrome. *Am J Med Genet* 1988;**29**:773-776.

357 Breslow NE, Beckwith JB. Epidemiological features of Wilms'tumor: results of the national Wilms'tumor study. *JNCI* 1982;**68**:429-436.

358 Matsunaga E. Genetics of Wilms' tumor. *Hum Genet* 1981;**57**:231-246.

359 Pastore G, CArli M, Lemerle J, et al. Epidemiological features of Wilms'tumor: results of studies by the international society of Peadiatric Oncology(SIOP). *Med Pediatr Oncol* 1988;**16**:7-11.

360 Bonaiti-Pellie C, Chompret A, Tournade MF, et al. Genetics and epidemiology of Wilms' tumor: The French Wilms'tumor study. *Med Pediatr Oncol* 1992;**20**:284-291.

361 Altura RA, Valentine M, Li H, et al. Identification of novel regions of deletion in familial Wilms' tumor by comparative genomic hybridization. *Cancer Res* 1996;**56**:3837-3841.

362 Rahman N, Arbour L, Tonin P, et al. Evidence for a familial Wilms'tumour gene (FWT1) on chromosome 17q12-q21. *Nat Genet* 1996;**13**:461-463.

363 Kaplinsky C, Ghahremani M, Frishberg Y, Rechavi G, Pelletier J. Familial Wilms' tumor associated with a WT1 zinc finger mutation. *Genomics* 1996;**38**:451-453.

364 Grundy P, Koufos A, Morgan K, Li FP, Meadows AT, Cavenee WK. Familial predisposition to Wilms'tumour does not map to the short arm of chromosome 11. *Nature* 1988;**336**:374-376.

365 Huff V, Compton DA, Chao LY, Strong LC, Geiser CF, Saunders GF. Lack of linkage of familial Wilms'tumour to chromosomal band 11p13. *Nature* 1988;**336**:377-378.

366 Huff V, Reeve AE, Leppert M, et al. Nonlinkage of 16q markers to familial predisposition to Wilm's tumor. *Cancer Res* 1992;**52**:6117-6120.

367 Schwartz CE, Haber DA, Stanton VP, Strong LC, Skolnick MH, Housman DE. Familial predisposition to Wilms tumor does not segregate with the WT1 gene. *Genomics* 1991;**10**:927-930.

368 Huff V, Amos CI, Douglass EC, et al. Evidence for genetic heterogeneity in familial Wilms' tumor. *Cancer Res* 1997;**57**:1859-1862.

369 Grundy RG, Pritchard J, Baraitser M, Risdon A, Robards M. Perlman and Wiedemann-Beckwith syndromes: two distinct conditions associated with Wilms'tumour. *Eur J Pediatr* 1992;**151**:895-898.

370 Neri G, Gurrieri F, Zanni G, Lin A. Clinical and molecular aspects of the Simpson-Golabi-Behmel syndrome. *Am J Med Genet* 1998;**79**:279-283.

371 Clericuzio CL, Johnson C. Screening for Wilms tumor in high-risk individuals. *Hematol Oncol Clin N Amer* 1995;**9**:1253

372 Sharpe CR, Franco EL. Etiology of Wilms' tumor. *Epidemiol Rev* 1995;**17**:415-432.

373 Maddock IR, Moran A, Maher ER, et al. A genetic register for von Hippel-Lindau disease. *J Med Genet* 1996;**33**:120-127.

374 Choyke PL, Walther MM, Glenn GM, et al. Imaging features of hereditary papillary renal cancers. *J Comput Assist Tomogr* 1997;**21**:737-741.

375 Sijmons RH, Kiemeney LALM, Witjes JA, Vasen HFA. Urinary tract cancer and hereditary nonpolyposis colorectal cancer: Risks and screening options. *J Urol* 1998;**160**:466-470.

376 Green DM, Breslow NE, Beckwith JB, Norkool P. Screening of children with hemihypertrophy, aniridia, and Bekwith-Wiedemann syndrome in patients with Wilms tumor: a report from the national Wilms tumor study. *Med Pediatr Oncol* 1993;**21**:188-192.

377 Schwartz AG, Yang P, Swanson GM. Familial risk of lung cancer among nonsmokers and their relatives. *Am J Epidemiol* 1996;**144**:554-562.

378 Sellers TA, Elston RC, Atwood LD, Rothschild H. Lung cancer histologic type and familly history of cancer. *Cancer* 1992;**69**:86-91.

379 Wu AH, Yu MC, Thomas DC, Pike MC, Henderson BE. Personal and family history of lung disease as risk factors for adenocarcinoma of the lung. *Cancer Res* 1988;**48**:7279-7284.

380 Ooi WL, Elston RC, Chen VW, Bailey-Wilson JE, Rothschild H. Increased familial risk for lung cancer. *J Natl Cancer Inst* 1986;**2**:217-222.

381 Samet JM, Humble CG, Pathak DR. Personal and family history of respiratory disease and lung cancer risk. *Am Rev Resp Dis* 1986;**134**:466-470.

382 Ambrosone CB, Rao U, Michalek AM, Cummings KM, Mettlin CJ. Lung cancer histologic types and family history of cancer. Analysis of histologic subtypes of 872 patients with primary lung cancer. *Cancer* 1993;**72**:1192-1198.

383 Shaw GL, Falk RT, Pickle LW, Mason TJ, Buffler PA. Lung cancer risk associated with cancer in relatives. *J Clin Epidemiol* 1991;**44**:429-437.

384 Wu AH, Fontham ETH, Reynolds P, et al. Family history of cancer and risk of lung cancer among lifetime nonsmoking women in the United States. *Am J Epidemiol* 1996;**143**:535-542.

385 Brownson RC, Alavanja MCR, Berger E, Chang JC. Family history of cancer and risk of lung cancer in lifetime non-smokers and long-term ex-smokers. *Int J Epidemiol* 1997;**26**:256-263.

386 Sellers TA, Chen PL, Potter JD, Bailey-Wilson JE, Rothschild H, Elston RC. Segregation analysis of smoking-associated malignancies: Evidence for Mendelian inheritance. *Am J Med Genet* 1994;**52**:308-314.

387 Braun MM, Caporaso NE, Page WF, Hoover RN. Genetic component of lung cancer: cohort studies of twins. *Lancet* 1994;**344**:440-443.

388 Gauderman WJ, Morrison JL, Carpenter JL, Thomas DC. Analysis of gene-smoking interaction in lung cancer. *Genet Epidemiol* 1997;**14**:199-214.

389 Yang P, Schwartz AG, McAllister AE, Aston CE, Swanson GM. Genetic analysis of families with nonsmoking lung cancer probands. *Genet Epidemiol* 1997;**14**:181-197.

390 Law MR. Genetic predisposition to lung cancer. *Br J Cancer* 1990;**61**:195-206.

391 Nakachi K, Imai K, Hayashi S, Wantanabe J, Kawajiri K. Genetic susceptibility to squamous cell carcinoma of the lung in relation to cigarette smoking dose. *Cancer Res* 1991;**51**:5177-5180.

392 Hirvonen A, Saarikoski ST, Linnainmaa K, et al. Glutathione S-transferase and N-acetyltransferase genotypes and asbestos-associated pulmonary disorders. *J Natl Cancer Inst* 1996;**88**:1853-1856.

393 Cascorbi I, Brockmoller J, Mrozikiewicz PM, Bauer S, Loddenkemper R, Roots I. Homozygous rapid arylamine N-acetyltransferase (NAT2) genotype as a susceptibility factor for lung cancer. *Cancer Res* 1996;**56**:3961-3966.

394 Hirvonen A, Pelin K, Tammilehto L, Karjalainen A, Mattson K, Linnainmaa K. Inherited GSTM1 and NAT2 defects as concurrent risk modifiers in asbestos-related human malignant mesothelioma. *Cancer Res* 1995;**55**:2981-2983.

395 Bouchardy C, Benhamou S, Dayer P. The effect of tobacco on lung cancer risk depends on CYP2D6 activity. *Cancer Res* 1996;**56**:251-253.

396 El-Zein RA, Zwischenberger JB, Abdel-Rahman SZ, Sankar AB, Au WW. Polymorphism of metabolizing genes and lung cancer histology: Prevalence of CYP2E1 in adenocarcinoma. *Cancer Lett* 1997;**112**:71-78.

397 El-Zein R, Conforti-Froes N, Au WW. Interactions between genetic predisposition and environmental toxicants for development of lung cancer. *Environ Mol Mutagen* 1997;**30**:196-204.

398 Ryberg D, Skaug V, Hewer A, et al. Genotypes of glutathione transferase M1 and P1 and their significance for lung DNA adduct levels and cancer risk. *Carcinogenesis* 1997;**18**:1285-1289.

399 Li FP, Lokich J, Lapey J, Neptune WB, Wilkins EW. Familial mesothelioma after intense asbestos exposure at home. *JAMA* 1978;**240**:467

400 Huncharek M, Kelsey K, Muscat J, Christiani D. Parental cancer and genetic predisposition in malignant pleural mesothelioma: A case-control study. *Cancer Lett* 1996;**102**:205-208.

401 Heineman EF, Bernstein L, Stark AD, Spirtas R. Mesothelioma, asbestos, and reported history of cancer in first-degree relatives. *Cancer* 1996;**77**:549-554.

402 Kleihues P, Schauble B, zur Hausen A, Esteve J, Ohgaki H. Tumors associated with p53 germline mutations: A synopsis of 91 families. *Am J Pathol* 1997;**150**:1-13.

403 Malkin D. p53 and the Li-Fraumeni syndrome. *Bba Rev Cancer* 1994;**1198**:197-213.

404 Nadav Y, Pastorino U, Nicholson AG. Multiple synchronous lung cancers and atypical adenomatous hyperplasia in Li-Fraumeni syndrome. *Histopathology* 1998;**33**:52-54.

405 Beaumont F, Jansen HM, Elema JD, ten Kate LP, Sluiter HJ. Simultaneous occurrence of pulmonary interstitial fibrosis and alveolar cell carcinoma in one family. *Thorax* 1981;**36**:252-258.

406 McKusick VA, Fisher AM. Congenital cystic disease of the lung with progressive pulmonary fibrosis and carcinomatosis. *Ann Intern Med* 1958;**48**:774-790.

407 Bonetti F, Chiodera P. Lymphangioleiomyomatosis and tuberous sclerosis: Where is the border? *Eur Resp J* 1996;**9**:399-401.

408 Kuhnen C, Muller KM, Khanavkar B, Nakhosteen JA. Pulmonary lymphangioleiomyomatosis in tuberous sclerosis. *Pathologe* 1994;**15**:354-357.

409 Nakhoul F, Kerner H, Levin M, Best LA, Better OS. Carcinoid tumor of the lung and type-1 multiple endocrine neoplasia associated with persistent hypercalcemia: A case report. *Miner Electrolyte Metab* 1994;**20**:107-111.

410 Sasaki M, Hakozaki H, Ishihara T. Cowden's disease with pulmonary hamartoma. *Intern Med* 1993;**32**:39-41.

411 Strauss GM, Gleason RE, Sugarbaker DJ. Screening for lung cancer - Another look; A different view. *Chest* 1997;**111**:754-768.

412 Mooney LA, Perera FP. Application of molecular epidemiology to lung cancer chemoprevention. *J Cell Biochem* 1996;**25S**:63-68.

413 Jefferies S, Eeles R, Goldgar D, AHern R, Henk JM, Gore M. The role of genetic factors in predisposition to squamous cell cancer of the head and neck. *Brit J Cancer* 1999;**79**:865-867.

414 Copper MP, Jovanovic A, Nauta JJP, et al. Role of genetic factors in the etiology of squamous cell carcinoma of the head and neck. *Arch Otolaryngol Head Neck Surg* 1995;**121**:157-160.

415 Foulkes WD, Brunet JS, Sieh W, Black MJ, Shenouda G, Narod SA. Familial risks of squamous cell carcinoma of the head and neck: Retrospective case-control study. *Br Med J* 1996;**313**:716-721.

416 Foulkes WD, Brunet J, Kowalski LP, Franco EL, Narod SA. Family history of cancer is a risk factor for squamous carcinoma of the head and neck: a case control study from Brazil. *Am J Hum Genet* 1995;**57**:A63

417 Wang YF, Chen CJ, Harris EL, et al. Complex segregation analysis of nasopharyngeal acrcinoma (NPC) in Taiwan (abstract). *Am J Hum Genet* 1995;**57**:A174

418 Sellers TA, Ooi WL, Elston RC, Chen VW, Bailey-Wilson JE, Rothschild H. Increased familial risk for non-lung cancer among relatives of lung cancer patients. *Am J Epidemiol* 1987;**2**:237-246.

419 Berkower AS, Biller HF. Head and neck cancer associated with Bloom's syndrome. *Laryngoscope* 1988;**98**:746-748.

420 Caceres-Rios H, Tamayo-Sanchez L, Duran-Mckinster C, de la Luz Orozco M, Ruiz-Maldonado R. Keratisis,Ichthyosis,and deafness (KID syndrome): review of the literature and proposal of a new terminology. *Pediatr Dermatol* 1996;**13**:105-113.

421 Yoshioka K, Kono T, Kitajima J, et al. Squamous cell carcinoma developing in epidermolysis bullosa dystrophica. *Int J Dermatol* 1991;**30**:718-721.

422 Kraemer KH. Lessons learned from xeroderma pigmentosum. *Photochem Photobiol* 1996;**63**:420-422.

423 Ambrosch P. Prevention of squamous cell carcinomas of the upper aerodigestive tract. In: Werner JA, Lippert BM, Rudert HH, eds. *Head and Neck Cancer - Advances in Basic Research*. 1000 AE Amsterdam: Elsevier Science Pub, 1996;629-634.

424 Huber MH, Hong WK. Biology and chemoprevention of head and neck cancer. *Curr Probl Cancer* 1994;**18**:81-140.

425 Lippman SM, Heyman RA, Kurie JM, Benner SE, Hong WK. Retinoids and chemoprevention: clinical and basic studies. *J Cell Biochem Suppl* 1995;**22: 1-10**:1-10.

426 Nguyen HN, Averette H, Janicek M. Ovarian carcinoma. A review of the significance of familial risk factors and the role of prophylactic oophorectomy in cancer prevention. *Cancer* 1994;**74**:545-555.

427 Verheijen RHM, Menko F, Kenemans P. Familiair ovariumcarcinoom. *Ned Tijdschr Geneeskd* 1994;**2**:63-66.

428 La Vecchia C, Parazzini F, Negri E, Del Pino D, Franceschi S. Family history and risk of ovarian cancer. *Int J Cancer* 1996;**67**:903-904.

429 Narod SA, Madlensky L, Bradley L, et al. Hereditary and familial ovarian cancer in southern Ontario. *Cancer* 1994;**74**:2341-2346.

430 Kerber RA, Slattery ML. The impact of family history on ovarian cancer risk: The Utah population database. *Arch Intern Med* 1995;**155**:905-912.

431 Claus EB, Schwartz PE. Familial ovarian cancer: Update and clinical applications. *Cancer* 1995;**76**:1998-2003.

432 Auranen A, Pukkala E, Makinen J, Sankila R, Grenman S, Salmi T. Cancer incidence in the first-degree relatives of ovarian cancer patients. *Br J Cancer* 1996;**74**:280-284.

433 Purdie D, Green A, Bain C, et al. Reproductive and other factors and risk of epithelial ovarian cancer: An Australian case-control study. *Int J Cancer* 1995;**62**:678-684.

434 Schildkraut JM, Thompson WD. Familial ovarian cancer: a population-based case-control study. *Am J Epidemiol* 1988;**3**:456-466.

435 Amos CI, Struewing JP. Genetic epidemiology of epithelial ovarian cancer. *Cancer* 1993;**71**:566-572.

436 Stratton JF, Pharoah P, Smith SK, Easton D, Ponder BAJ. A systematic review and meta-analysis of family history and risk of ovarian cancer. *Br J Obstet Gynaecol* 1998;**105**:493-499.

437 Steinberg KK, Pernarelli JM, Marcus M, Khoury MJ, Schildkraut JM, Marchbanks PA. Increased risk for familial ovarian cancer among Jewish women: A population-based case-control study. *Genet Epidemiol* 1998;**15**:51-59.

438 Foulkes WD, Narod SA. Ovarian cancer risk and family history. *Lancet* 1997;**349**:878

439 Amos CI, Shaw GL, Tucker MA, Hartge P. Age at onset for familial epithelial ovarian cancer. *JAMA* 1992;**268**:1896-1899.

440 Goldberg JM, Piver MS, Jishi MF, Blumenson L. Age at onset of ovarian cancer in women with a strong family history of ovarian cancer . *Gynecol Oncol* 1997;**66**:3-9.

441 Cramer DW, Hutchison GB, Welch WR, Scully RE, Ryan KJ. Determinants of ovarian cancer risk. I. Reproductive experiences and family history. *J Natl Cancer Inst* 1983;**71**:711-716.

442 Narod SA. Genetics of breast and ovarian cancer. *Br Med Bull* 1994;**50**:656-676.

443 Piver MS, Baker TR, Jishi MF, et al. Familial ovarian cancer. A report of 658 families from the Gilda Radner Familial Ovarian Cancer Registry 1981-1991. *Cancer* 1993;**71**:582-588.

444 Longy M, Toulouse C, Mage P, Chauvergne M, Trojani M. Familial cluster of ovarian small cell carcinoma: A new mendelian entity? *J Med Genet* 1996;**33**:333-335.

445 Jackson SM. Ovarian dysgerminoma in three generations? *J Med Genet* 1967;**4**:112-113.

446 Dumont-Herskowitz RA, Safaii HS, Senior B. Ovarian fibromata in four successive generations. *J Pediat* 1978;**4**:621-624.

447 Piver MS, Goldberg JM, Tsukada Y, Mettlin CJ, Jishi MF, Natarajan N. Characteristics of familial ovarian cancer: a report of the first 1,000 families in the Gilda Radner Familial Ovarian Cancer Registry. *Eur J Gynaecol Oncol* 1996;**17**:169-176.

448 Schildkraut JM, Thompson WD. Relationship of epithelial ovarian cancer to other malignancies within families. *Genet Epidemiol* 1988;**5**:355-367.

449 Shushan A, Paltiel O, Gordon L, Schenker JG. Ovarian cancer of low malignant potential is not associated with positive familial history. *Am J Obstet Gynecol* 1996;**175**:507-508.

450 Rader JS, Neuman RJ, Brady J, et al. Cancer among first-degree relatives of probands with invasive and borderline ovarian cancer. *Obstet Gynecol* 1998;**92**:589-595.

451 Stratton JF, Gayther SA, Russell P, et al. Contribution of BRCA1 mutations to ovarian cancer. *N Engl J Med* 1997;**336**:1125-1130.

452 Whittemore AS, Gong G, Itnyre J. Prevalence and contribution of BRCA1 mutations in breast cancer and ovarian cancer: Results from three U.S. population-based case-control studies of ovarian cancer. *Am J Hum Genet* 1997;**60**:496-504.

453 Berchuck A, Heron KA, Carney ME, et al. Frequency of germline and somatic BRCA1 mutations in ovarian cancer. *Clin Cancer Res* 1998;**4**:2433-2437.

454 Narod SA, Goldgar D, Cannonalbright L, et al. Risk modifiers in carriers of BRCA1 mutations. *Int J Cancer* 1995;**64**:394-398.

455 Narod S, Tonin P, Lynch H, Watson P, Feunteun J, Lenoir G. Histology of BRCA1-associated ovarian tumours. *Lancet* 1994;**343**:236

456 Shushan A, Abeliovich D, Peretz T, Weinberg N, Paltiel O. BRCA1 mutations in women with ovarian cancer. *JAMA* 1997;**277**:963

457 Modan B, Friedman E, Hirsh-Yechezkel G. BRCA1 mutations in women with ovarian cancer - Reply. *JAMA* 1997;**277**:963

458 Gotlieb WH, Freidman E, Bar-Sade RB, et al. Rates of Jewish ancestral mutations in BRCA1 and BRCA2 in borderline ovarian tumors. *J Natl Cancer Inst* 1998;**90**:995-1000.

459 Lu KH, Cramer DW, Muto MG, Li EY, Niloff J, Mok SC. A population-based study of BRCA1 and BRCA2 mutations in Jewish women with epithelial ovarian cancer. *Obstet Gynecol* 1999;**93**:34-37.

460 Frank TS, Manley SA, Olopade OI, et al. Sequence analysis of BRCA1 and BRCA2: Correlation of mutations with family history and ovarian cancer risk. *J Clin Oncol* 1998;**16**:2417-2425.

461 Rubin SC, Benjamin I, Behbakht K, et al. Clinical and pathological features of ovarian cancer in women with germ-line mutations of BRCA1. *NEJM* 1996;**335**:1413-1416.

462 Aida H, Takakuwa K, Nagata H, et al. Clinical features of ovarian cancer in Japanese women with germ-line mutations of BRCA1. *Clin Cancer Res* 1998;**4**:235-240.

463 Johannsson O, Ranstam J, Borg A, Olsson H. BRCA1 mutations and survival in women with ovarian cancer. *N Engl J Med* 1997;**336**:1255-1256.

464 Brunet JS, Narod SA, Tonin P, Foulkes WD. BRCA1 mutations and survival in women with ovarian cancer. *N Engl J Med* 1997;**336**:1256

465 Pharoah PDP, Easton DF, Stockton DL, Gayther S, Ponder BAJ. Survival in familial, BRCA1-associated, and BRCA2-associated epithelial ovarian cancer. *Cancer Res* 1999;**59**:868-871.

466 Scully RE. Sex cord tumor with annular tubules a distinctive ovarian tumor of the Peutz-Jeghers syndrome. *Cancer* 1970;**2**:1107-1121.

467 Hecht F, Hecht BK. Cancer in ataxia-telangiectasia patients: 10th anniversary article. *Cancer Genet Cytogenet* 1990;**46**:9-19.

468 Moorthy AV, Chesney RW, Lubinsky M. Chronic renal failure and XY gonadal dysgenesis: "Frasier" syndrome--a commentary on reported cases. *Am J Med Genet Suppl* 1987;**3: 297-302**:297-302.

469 Mueller RF. The Denys-Drash syndrome. *J Med Genet* 1994;**31**:471-477.

470 Dorum A, Kristensen GB, Abeler VM, Trope CG, Moller P. Early detection of familial ovarian cancer. *Eur J Cancer* 1996;**32A**:1645-1651.

471 Bourne TH, Whitehead MI, Campbell S, Royston P, Bhan V, Collins WP. Ultrasound screening for familial ovarian cancer. *Gynecol Oncol* 1991;**43**:92-97.

472 Muto MG, Cramer DW, Brown DL, et al. Screening for ovarian cancer: the preliminary experience of a familial ovarian cancer center. *Gynecol Oncol* 1993;**51**:12-20.

473 Jacobs I. Genetic, Biochemical, and multimodal approaches to screening for ovarian cancer. *Gynecol Oncol* 1994;**55**:S22-S27.

474 Karlan BY, Raffel LJ, Crvenkovic G, et al. A multidisciplinary approach to the early detection of ovarian carcinoma: Rationale, protocol design, and early results. *Am J Obstet Gynecol* 1993;**169**:494-501.

475 Bourne TH, Campbell S, Reynolds KM, et al. Screening for early familial ovarian cancer with transvaginal ultrasonography and colour blood flow imaging. *BMJ* 1993;**306**:1025-1029.

476 Greely HT. Genetic testing for cancer susceptibility: challenges for creators of practice guidelines. *Oncology Huntingt* 1997;**11**:171-176.

477 Kerlikowske K, Brown JS, Grady DG. Should women with familial ovarian cancer undergo prophylactic oophorectomy ? *Obstet Gynecol* 1992;**80**:700-707.

478 Lynch HT, Albano WA, Lynch JF, Lynch PM, Campbell A. Surveillance and management of patients at high genetic risk for ovarian carcinoma. *Obstet Gynecol* 1982;**5**:589-596.

479 Daly MB, Lerman C. Ovarian cancer risk counseling: a guide for the practitioner. *Oncology* 1993;**11**:27-34.

480 Piver MS, Jishi MF, Tsukada Y, Nava G. Primary peritoneal carcinoma after prophylactic oophorectomy in women with a family history of ovarian cancer. A report of the Gilda Radner Familial Ovarian Cancer Registry. *Cancer* 1993;**71**:2751-2755.

481 Tobacman JK, Greene MH, Tucker MA, Costa J, Kase R, Fraumeni jr JF. Intra-abdominal carcinomatosis after prophylactic oophorectomy in ovarian-cancer-prone families. *Lancet* 1982;**II**:795-797.

482 Struewing JP, Watson P, Easton DF, Ponder BAJ, Lynch HT, Tucker MA. Prophylactic oophorectomy in inherited breast/ovarian cancer families. *J Natl Cancer Inst Mono* 1995;**17**:33-35.

483 Schorge JO, Muto MG, Welch WR, et al. Molecular evidence for multifocal papillary serous carcinoma of the peritoneum in patients with germline BRCA1 mutations. *J Natl Cancer Inst* 1998;**90**:841-845.

484 Eisen A, Weber BL. Primary peritoneal carcinoma can have multifocal origins: Implications for prophylactic oophorectomy. *J Natl Cancer Inst* 1998;**90**:797-799.

485 Harlap S. The epidemiology of ovarian cancer. In: Hoskins WJ, Markman M, eds. *Cancer of the Ovary*. New York: Raven press, 1993;79-93.

486 Narod SA, Risch H, Moslehi R, et al. Oral contraceptives and the risk of hereditary ovarian cancer. *N Engl J Med* 1998;**339**:424-428.

487 Rubin SC. Chemoprevention of hereditary ovarian cancer. *N Engl J Med* 1998;**339**:469-471.

488 Lynch HT, Smyrk T, Kern SE, et al. Familial pancreatic cancer: A review. *Semin Oncol* 1996;**23**:251-275.

489 Flanders TY, Foulkes WD. Pancreatic adenocarcinoma: epideliology and genetics. *J Med Genet* 1996;**33**:889-898.

490 Lumadue JA, Griffin CA, Osman M, Hruban RH. Familial pancreatic cancer and the genetics of pancreatic cancer. *Surg Clin North Am* 1995;**75**:845

491 Dergham ST, Dugan MC, Arlauskas P, et al. Relationship of family cancer history to the expression of p53, p21(WAF-1), HER-2/neu, and K-ras mutation in pancreatic adenocarcinoma. *Int J Pancreatol* 1997;**21**:225-234.

492 Fernandez E, La Vecchia C, D'Avanzo B, Negri E, Franceschi S. Family history and the risk of liver, gallbladder, and pancreatic cancer. *Cancer Epidemiol Biomarkers Prev* 1994;**3**:209-212.

493 Falk RT, Pickle LW, Fontham ET, Correra P, Fraumeni jr JF. Life-style risk factors for pancreatic cancer in Louisiana: a case-control study. *Am J Epidemiol* 1988;**128**:324-336.

494 Tulinius H, Olafsdottir GH, Sigvaldason H, Tryggvadottir L, Bjarnadottir K. Neoplastic diseases in families of breast cancer patients. *J Med Genet* 1994;**31**:618-621.

495 Ehrenthal D, Haeger L, Griffin T, Compton C. Familial pancreatic adenocarcinoma in three generations. A case report and review of the literature. *Cancer* 1987;**59**:1661-1664.

496 Burke W, Bennett RL, Schmidt R, Dellinger P, Evans JP. Autosomal dominant transmission of pancreatic cancer with diabetes and exocrine insuficiency in a large kindred . *Am J Hum Genet* 1992;**51**:A50

497 Evans JP, Burke W, Chen R, et al. Familial pancreatic adenocarcinoma: Association with diabetes and early molecular diagnosis. *J Med Genet* 1995;**32**:330-335.

498 Scott-Conner CEH, Hausmann M, Hall TJ, Skelton DS, Anglin BL, Subramony C. Familial juvenile Polyposis: Patterns of recurrence and implications for surgical management. *J Amer Coll Surgeons* 1995;**181**:407-413.

499 Tsongalis GJ, Faber G, Dalldorf FG, Friedman KJ, Silverman LM, Yankaskas JR. Association of pancreatic adenocarcinoma, mild lung disease, and Delta F508 mutation in a cystic fibrosis patient. *Clin Chem* 1994;**40**:1972-1974.

500 Goggins M, Schutte M, Lu J, et al. Germline BRCA2 gene mutations in patients with apparently sporadic pancreatic carcinomas. *Cancer Res* 1996;**56**:5360-5364.

501 Ozcelik H, Schmocker B, DiNicola N, et al. Germline BRCA2 6174delT mutations in Ashkenazi Jewish pancreatic cancer patients. *Nat Genet* 1997;**16**:17-18.

502 Padberg B, Schroder S, Capella C, Frilling A, Kloppel G, Heitz PU. Multiple endocrine neoplasia type 1 (MEN 1) revisited. *Virchows Archiv* 1995;**426**:541-548.

503 Chanson P, Cadiot G, Murat A. Management of patients and subjects at risk for multiple endocrine neoplasia type 1: MEN 1. *Horm Res* 1997;**47**:211-220.

504 Cussenot O, Valeri A, Meria P, et al. Genetic factors in prostate cancer (French). *Pathol Biol* 1996;**44**:737-743.

505 Ghadirian P, Howe GR, Hislop TG, Maisonneuve P. Family history of prostate cancer: A multi-center case-control study in Canada. *Int J Cancer* 1997;**70**:679-681.

506 Woolf CM. An investigation of the familial aspects of carcinoma of the prostate. *Cancer* 1960;**13**:739-744.

507 Key T. Risk factors for prostate cancer. *Cancer Surv* 1995;**2363-77**:-77.

508 Pienta KJ, Esper PS. Risk factors for prostate cancer. *Am Coll Phys* 1993;**118**:793-803.

509 Carter BS, Steinberg GD, Beaty TH, Childs B, Walsh PC. Familial risk factors for prostate cancer. *Cancer Surv* 1991;**11**:5-13.

510 Giovannucci E. How is individual risk for prostate cancer assessed? *Hematol Oncol Clin N Amer* 1996;**10**:537

511 Walsh PC, Partin AW. Family history facilitates the early diagnosis of prostate carcinoma. *Cancer* 1997;**80**:1871-1874.

512 Steinberg GD, Carter BS, Beaty TH, Childs B, Walsh PC. Family history and the risk of prostate cancer. *The Prostate* 1990;**17**:337-347.

513 Narod SA, Dupont A, Cusan L, et al. The impact of family history on early detection of prostate cancer. *Nat Med* 1995;**1**:99-101.

514 Hayes RB, Liff JM, Pottern LM, et al. Prostate cancer risk in US blacks and whites with a family history of cancer. *Int J Cancer* 1995;**60**:361-364.

515 Spitz MR, Currier RD, Fueger JJ, Babaian RJ, Newell GR. Familial patterns of prostate cancer: a case-control analysis. *J Urol* 1991;**146**:1305-1307.

516 Nguyen D, Wachtel M, Payami H. Risk of prostate cancer, breast cancer and ovarian cancer in parents and siblings of prostate cancer patients (abstract). *Am J Hum Genet* 1995;**57**:A74

517 Lesko SM, Rosenberg L, Shapiro S. Family history and prostate cancer risk. *Am J Epidemiol* 1996;**144**:1041-1047.

518 Gronberg H, Damber L, Damber JE. Familial prostate cancer in Sweden: A nationwide register cohort study. *Cancer* 1996;**77**:138-143.

519 Keetch DW, Rice JP, Suarez BK, Catalona WJ. Familial aspects of prostate cancer: a case control study. *J Urol* 1995;**154**:2100-2102.

520 Meikle AW, Smith JA, West DW. Familial factors affecting prostatic cancer risk and plasma sex-steroid levels. *Prostate* 1985;**6**:121-128.

521 Whittemore AS, Wu AH, Kolonel LN, et al. Family history and prostate cancer risk in black, white, and Asian men in the United States and Canada. *Am J Epidemiol* 1995;**141**:732-740.

522 Bratt O, Kristoffersson U, Lundgren R, Olsson H. The risk of malignant tumours in first-degree relatives of men with early onset prostate cancer: A population-based cohort study. *Eur J Cancer* 1997;**33**:2237-2240.

523 Gronberg H, Damber L, Damber JE. Studies of genetic factors in prostate cancer in a twin population. *J Urol* 1994;**152**:1484-1487.

524 Page WF, Braun MM, Partin AW, Caporaso N, Walsh P. Heredity and prostate cancer: A study of World War II veteran twins. *Prostate* 1997;**33**:240-245.

525 Rodriguez C, Calle EE, Tatham LM, et al. Family history of breast cancer as a predictor for fatal prostate cancer. *Epidemiology* 1998;**9**:525-529.

526 Carter BS, Beaty TH, Steinberg GD, Childs B, Walsh PC. Mendelian inheritance of familial prostate cancer. *Proc Natl Acad Sci USA* 1992;**89**:3367-3371.

527 Bishop DT, Kiemeney LA. Family studies and the evidence for genetic susceptibility to prostate cancer. *Sem Cancer Biol* 1997;**8**:45-51.

528 Schaid DJ, McDonnell SK, Blute ML, Thibodeau SN. Evidence for autosomal dominant inheritance of prostate cancer. *Am J Hum Genet* 1998;**62**:1425-1438.

529 Gronberg H, Damber L, Damber JE, Iselius L. Segregation analysis of prostate cancer in Sweden: Support for dominant inheritance. *Am J Epidemiol* 1997;**146**:552-557.

530 Smith JR, Freije D, Carpten JD, et al. Major susceptibility locus for prostate cancer on chromosome 1 suggested by a genome-wide search. *Science* 1996;**274**:1371-1374.

531 Xu JF, Meyers D, Freije D, et al. Evidence for a prostate cancer susceptibility locus on the X chromosome. *Nat Genet* 1998;**20**:175-179.

532 Kupelian PA, Kupelian VA, Witte JS, Macklis R, Klein EA. Family history of prostate cancer in patients with localized prostate cancer: An independent predictor of treatment outcome. *J Clin Oncol* 1997;**15**:1478-1480.

533 Kupelian PA, Klein EA, Witte JS, Kupelian VA, Suh JH. Familial prostate cancer: A different disease? *J Urol* 1997;**158**:2197-2201.

534 Rodriguez C, Calle EE, Miracle-McMahill HL, et al. Family history and risk of fatal prostate cancer. *Epidemiology* 1997;**8**:653-657.

535 Hanus MC, Zagars GK, Pollack A. Familial prostate cancer: Outcome following radiation therapy with or without adjuvant androgen ablation. *Int J Radiat Oncol Biol Phys* 1999;**43**:379-383.

536 Gronberg H, Damber L, Tavelin B, Damber JE. No difference in survival between sporadic, familial and hereditary prostate cancer. *Br J Urol* 1998;**82**:564-567.

537 Carter BS, Bova GS, Beaty TH, et al. Hereditary prostate cancer: epidemiologic and clinical features. *J Urol* 1993;**150**:797-802.

538 Bastacky SI, Wojno KJ, Walsh PC, Carmichael MJ, Epstein JI. Pathological features of hereditary prostate cancer. *J Urol* 1995;**153**:987-992.

539 Aprikian AG, Bazinet M, Plante M, et al. Family history and the risk of prostatic carcinoma in a high risk group of urological patients. *J Urol* 1995;**154**:404-406.

540 Keetch DW, Humphrey PA, Smith DS, Stahl D, Catalona WJ. Clinical and pathological features of hereditary prostate cancer. *J Urol* 1996;**155**:1841-1843.

541 Gronberg H, Isaacs SD, Smith JR, et al. Characteristics of prostate cancer in families potentially linked to the hereditary prostate cancer 1 (HPC1) locus. *JAMA* 1997;**278**:1251-1255.

542 Langston AA, Stanford JL, Wicklund KG, Thompson JD, Blazej RG, Ostrander EA. Germ-line BRCA1 mutations in selected men with prostate cancer. *Am J Hum Genet* 1996;**58**:881-885.

543 Thorlacius S, Sigurdsson S, Bjarnadottir H, et al. Study of a single BRCA2 mutation with high carrier frequency in a small population. *Am J Hum Genet* 1997;**60**:1079-1084.

544 Eeles R, Mc.Whorter W, Smith D, et al. Prostate specific antigen in relatives of cases in high risk Utah prostate cancer kindreds: an argument for targeted prostate cancer screening (abstract). *Am J Hum Genet* 1995;**57**:A294

545 Hernandez AD, Mcwhorter WP, Meikle W, et al. A prospective study of prostate cancer in high risk families (abstract). *J Urol* 1992;**147**:451A

546 Jacobson JO. Can screening for early-stage prostate cancer be rationalized? *Hematol Oncol Clin N Amer* 1996;**10**:549

547 Kiemeney LALM, Witjes JA, Hendrikx AJM, Kil PJM, Vasen HFA. Erfelijk prostaatcarcinoom. *Ned Tijdschr Geneeskd* 1996;**140**:1068-1072.

548 Mclellan DL, Norman RW. Hereditary aspects of prostate cancer. *Can Med Assoc J* 1995;**153**:895-900.

549 Brawley OW, Thompson IM. The chemoprevention of prostate cancer and the Prostate Cancer Prevention Trial. *Cancer Treat Res* 1996;**88: 189-200**:189-200.

550 Thompson IM, Coltman CA, Jr., Crowley J. Chemoprevention of prostate cancer: the Prostate Cancer Prevention Trial. *Prostate* 1997;**33**:217-221.

551 Liu TP, Soong SJ. Epidemiology of malignant melanoma. *Surg Clin North Am* 1996;**76**:1205

552 Grange F, Chompret A, Guilloudbataille M, et al. Comparison between familial and nonfamilial melanoma in France. *Arch Dermatol* 1995;**131**:1154-1159.

553 Briollais L, Chompret A, Guilloud-Bataille M, Feingold N, Avril MF, Demenais F. Genetic and epidemiological risk factors for a malignant melanoma- predisposing phenotype: The great number of nevi. *Genet Epidemiol* 1996;**13**:385-402.

554 Breitbart M, Metneki J, Weichenthal M, et al. A study of Hungarian twins involving the influence of genetic and environmental factors on benign, melanocytic lesions. *Eur J Dermatol* 1996;**6**:548-551.

555 Aitken JF, Duffy DL, Green A, Youl P, Maclennan R, Martin NG. Heterogeneity of melanoma risk in families of melanoma patients. *Am J Epidemiol* 1994;**140**:961-973.

556 Ford D, Bliss JM, Swerdlow AJ, et al. Risk of cutaneous melanoma associated with a family history of the disease. The International Melanoma Analysis Group (IMAGE). *Int J Cancer* 1995;**62**:377-381.

557 Cannon-Albright LA, Kamb A, Skolnick M. A review of inherited predisposition to melanoma. *Semin Oncol* 1996;**23**:667-672.

558 Lynch HT, Fusaro RM, Danes BS, Kimberling WJ, Lynch JF. A review of hereditary malignant melanoma including biomarkers in familial atypical multiple mole melanoma syndrome. *Cancer Genet Cytogenet* 1983;**8**:325-358.

559 Bergman W, Gruis NA, Sandkuijl LA, Frants RR. Genetics of seven Dutch familial atypical multiple mole melanoma syndrome families: A review of linkage results including chromosomes 1 and 9. *J Invest Dermatol* 1994;**103**:S122-S125.

560 Walker GJ, Nancarrow DJ, Walters MK, Palmer JM, Weber JL, Hayward NK. Linkage analysis in familial melanoma kindreds to markers on chromosome 6p. *Int J Cancer* 1994;**59**:771-775.

561 Novakovic B, Clark WH, Fears TR, Fraser MC, Tucker MA. Melanocytic nevi, dysplastic nevi, and malignant melanoma in children from melanoma-prone families. *J Am Acad Dermatol* 1995;**33**:631-636.

562 Hussussian CJ, Struewing JP, Goldstein AM, et al. Germline p16 mutations in familial melanoma. *Nat Genet* 1994;**8**:15-21.

563 NIH Consensus Conference. Diagnosis and treatment of early melanoma. *JAMA* 1992;**268**:1314-1319.

564 Zuo L, Weger J, Yang QB, et al. Germline mutations in the p16(INK4a) binding domain of CDK4 in familial melanoma. *Nat Genet* 1996;**12**:97-99.

565 Fitzgerald MG, Harkin DP, Silva-Arrieta S, et al. Prevalence of germ-line mutations in p16, p19ARF, and CDK4 in familial melanoma: Analysis of a clinic-based population. *Proc Natl Acad Sci USA* 1996;**93**:8541-8545.

566 Goldstein AM, Tucker MA. Screening for CDKN2A mutations in hereditary melanoma. *J Natl Cancer Inst* 1997;**89**:676-678.

567 Cannon-Albright LA, Meyer LJ, Goldgar DE, et al. Penetrance and expressivity of the chromosome 9p melanoma susceptibility locus (MLM). *Cancer Res* 1994;**54**:6041-6044.

568 Battistutta D, Palmer J, Walters M, Walker G, Nancarrow D, Hayward N. Incidence of familial melanoma and MLM2 gene. *Lancet* 1994;**344**:1607-1608.

569 Kiemeney L. Frequency of familial melanoma and MLM2 gene. *Lancet* 1995;**345**:581

570 Bataille V, Hiles R, Bishop JAN. Retinoblastoma, melanoma and the atypical mole syndrome. *Br J Dermatol* 1995;**132**:134-138.

571 Witkop CJ, Jr. Albinism. *Adv Hum Genet* 1971;**2**:61-142.

572 Goto M, Miller RW, Ishikawa Y, Sugano H. Excess of rare cancers in Werner syndrome (adult progeria). *Cancer Epidemiol Biomarkers Prev* 1996;**5**:239-246.

573 Kraemer KH, DiGiovanna JJ, Moshell AN, Tarone RE, Peck GL. Prevention of skin cancer in xeroderma pigmentosum with the use of oral isotretinoin. *N Engl J Med* 1988;**318**:1633-1637.

574 Kraemer KH, DiGiovanna JJ, Peck GL. Chemoprevention of skin cancer in xeroderma pigmentosum. *J Dermatol* 1992;**19**:715-718.

575 Maia M, Proenca NG, de Moraes JC. Risk factors for basal cell carcinoma: a case-control study. *Rev Saude Publica* 1995;**29**:27-37.

576 Gellin GA, Kope AW, Garfinkel L. Basal cell epithelioma: A controlled study of associated factors. *Arch Dermatol* 1965;**91**:38-45.

577 Wallberg P, Kaaman T, Lindberg M. Multiple basal cell carcinoma - A clinical evaluation of risk factors. *Acta Derm Venereol Stockh* 1998;**78**:127-129.

578 Armstrong BK, Kricker A. Epidemiology of sun exposure and skin cancer. *Cancer Surv* 1996;**26**:133-153.

579 Hogan DJ, To T, Gran L, Wong D, Lane PR. Risk factors for basal cell carcinoma. *Int J Dermatol* 1989;**28**:591-594.

580 Hogan DJ, Lane PR, Gran L, Wong D. Risk factors for squamous cell carcinoma of the skin in Saskatchewan, Canada. *J Dermatol Sci* 1990;**1**:97-101.

581 Gamble JF, Lerman SE, Holder WR, Nicolich MJ, Yarborough CM. Physician-based case-control study of non-melanoma skin cancer in Baytown, Texas. *Occup Med Oxf* 1996;**46**:186-196.

582 Van Hees CLM, Van Duinen CM, Bruijin JA, Vermeer BJ. Malignant eccrine poroma in a patient with Rothmund-Thomson syndrome. *Br J Dermatol* 1996;**134**:813-815.

583 Kidd A, Carson L, Gregory DW, et al. A Scottish family with Bazex-Dupre-Christol syndrome: Follicular atrophoderma, congenital hypotrichosis, and basal cell carcinoma. *J Med Genet* 1996;**33**:493-497.

584 Michaelsson G, Westermark P. The rombo syndrome: a familial disorder with vermiculate atrophoderma, milia, hypotrichosis, trichoepeitheliomas, basal cell carcinomas and peripheral vasodilation with cyanosis. *Acta Derm Venereol Stockh* 1981;**61**:497-503.

585 Hamm H, Traupe H, Brocker EB, Schubert H, Kolde G. The scleroatrophic syndrome of Huriez: A cancer-prone genodermatosis. *Br J Dermatol* 1996;**134**:512-518.

586 Sirinavin C, Trowbridge AA. Dyskeratosis congenita: clinical features and genetic aspects: report of a family and review of the literature. *J Med Genet* 1975;**12**:339-354.

587 Grob JJ, Breton A, Bonafe JL, Sauvan Ferdani M, Bonerandi JJ. Keratitis, ichthyosis, and deafness (KID) syndrome. Vertical transmission and death from multiple squamous cell carcinomas. *Arch Dermatol* 1987;**123**:777-782.

588 Madariaga J, Fromowitz F, Phillips M, Hoover HC, Jr. Squamous cell carcinoma in congenital ichthyosis with deafness and keratitis. A case report and review of the literature. *Cancer* 1986;**57**:2026-2029.

589 Ferguson-Smith MA, Wallace DC, James Z, Renwick JH. Multiple self-healing squamous epithelioma. *Birth Defects* 1971;**7**:157-163.

590 McGrath JA, Schofield OMV, Mayou BJ, McKee PH, Eady RAJ. Epidermolysis bullosa complicated by squamous cell carcinoma: report of 10 cases. *J Cutan Pathol* 1992;**19**:116-123.

591 Swensson O, Christophers E. Generalized atrophic benign epidermolysis bullosa in 2 siblings complicated by multiple squamous cell carcinomas. *Arch Dermatol* 1998;**134**:199-203.

592 Schamroth JM, Zlotogorski A, Gilead L. Porokeratosis of Mibelli. Overview and review of the literature. *Acta Derm Venereol* 1997;**77**:207-213.

593 Schwartz RA, Torre DP. The Muir-Torre syndrome: A 25-year retrospect. *J Am Acad Dermatol* 1995;**33**:90-104.

594 Biggs PJ, Wooster R, Ford D, et al. Familial cylindromatosis (turban tumour syndrome) gene localised to chromosome 16q12-q13: Evidence for its role as a tumour suppressor gene. *Nat Genet* 1995;**11**:441-443.

595 Gerretsen AL, Beemer FA, Deenstra W, Hennekam FAM, Van Vloten WA. Familial cutaneous cylindromas: Investigations in five generations of a family. *J Am Acad Dermatol* 1995;**33**:199-206.

596 Mallory SB. Cowden syndrome (multiple hamartoma syndrome). *Dermatol Clin* 1995;**13**:27-31.

597 Listernick R, Charrow J. Neurofibromatosis typr 1 in childhood. *J Pediat* 1990;**116**:845-853.

598 Bundey S, Evans K. Tuberous sclerosis: a genetic study. *J Neurol Neurosurg Psychiatry* 1969;**32**:591-603.

599 McCauley RG, Leonidas JC, Bartoshesky LE. Blue rubber bleb nevus syndrome. *Radiology* 1979;**133**:375-377.

600 Levine N, Moon TE, Cartmel B, et al. Trial of retinol and isotretinoin in skin cancer prevention: a randomized, double-blind, controlled trial. Southwest Skin Cancer Prevention Study Group. *Cancer Epidemiol Biomarkers Prev* 1997;**6**:957-961.

601 Zanghieri G, Di Gregorio C, Sachetti C, et al. Familial occurrence of gastric cancer in the 2-year experience of a population-based registry. *Cancer* 1990;**66**:2047-2051.

602 Macklin MT. Inheritance of cancer of the stomach and large intestine in man. *J Natl Cancer Inst* 1960;**24**:551-571.

603 Ponz de leon M. *Familial and hereditary tumors*. Berlin: Springer-Verlag, 1994;

604 Fuchs CS, Mayer RJ. Medical progress: Gastric carcinoma. *N Engl J Med* 1995;**333**:32-41.

605 Lehtola J. Family study of gastric carcinoma; with special reference to histological types. *Scand J Gastroenterol* 1978;**Suppl.50**:1-54.

606 La Vecchia C, Negri E, Franceschi S, Gentile F. Family history and the risk of stomach and colorectal cancer. *Cancer* 1992;**70**:50-55.

607 Nagase H, Ogino K, Yoshida I, et al. Family history-related risk of gastric cancer in Japan: A hospital- based case-control study. *Jpn J Cancer Res* 1996;**87**:1025-1028.

608 Inoue M, Tajima K, Yamamura Y, et al. Family history and subsite of gastric cancer: Data from a case-referent study in Japan. *Int J Cancer* 1998;**76**:801-805.

609 Kikuchi S, Nakajima T, Nishi T, et al. Association between family history and gastric carcinoma among young adults. *Jpn J Cancer Res* 1996;**87**:332-336.

610 Morita M, Kuwano H, Baba H, et al. Multifocal occurrence of gastric carcinoma in patients with a family history of gastric carcinoma. *Cancer* 1998;**83**:1307-1311.

611 Gasbarrini G, Pretolani S, Bonvicini F, et al. A population based study of Helicobacter pylori infection in a European country: The San Marino study. Relations with gastrointestinal diseases. *Gut* 1995;**36**:838-844.

612 Novakovic B, Goldstein AM, Wexler LH, Tucker MA. Increased risk for neuroectodermal tumors and stomach cancer in relatives of patients with Ewing's sarcoma. *J Natl Cancer Inst* 1994;**86**:1702-1706.

613 Guilford P, Hopkins J, Harraway J, et al. E-cadherin germline mutations in familial gastric cancer. *Nature* 1998;**392**:402-405.

614 Gayther SA, Gorringe KL, Ramus SJ, et al. Identification of germ-line E-cadherin mutations in gastric cancer families of European origin. *Cancer Res* 1998;**58**:4086-4089.

615 Richards FM, McKee SA, Cole TRP, et al. Germline e-cadherin gene mutations in familial gastric cancer. *J Med Genet* 1998;**35**:S24

616 Peddanna N, Holt S, Verma RS. Genetics of gastric cancer. *Anticancer Res* 1995;**15**:2055-2064.

617 Aarnio M, Salovaara R, Aaltonen LA, Mecklin JP, Jarvinen HJ. Features of gastric cancer in hereditary non-polyposis colorectal cancer syndrome. *Int J Cancer* 1997;**74**:551-555.

618 Keller G, Rotter M, Vogelsang H, et al. Microsatellite instability in adenocarcinomas of the upper gastrointestinal tract: Relation to clinicopathological data and family history. *Am J Pathol* 1995;**147**:593-600.

619 Keller G, Grimm V, Vogelsang H, et al. Analysis for microsatellite instability and mutations of the DNA mismatch repair gene hMLH1 in familial gastric cancer. *Int J Cancer* 1996;**68**:571-576.

620 Akiyama Y, Nagasaki H, Nihei Z, et al. Frequent microsatellite instabilities and analyses of the related genes in familial gastric cancers. *Jpn J Cancer Res* 1996;**87**:595-601.

621 Chong JM, Fukayama M, Hayashi Y, et al. Microsatellite instability in the progression of gastric carcinoma. *Cancer Res* 1994;**54**:4595-4597.

622 Cunningham-Rundles C. Clinical and immunologic analyses of 103 patients with common variable immunodeficiency. *J Clin Immunol* 1989;**9**:22-33.

623 Filipovich AH, Mathur A, Kamat D, Kersey JH, Shapiro RS. Lymphoproliferative disorders and other tumors complicating immunodeficiencies. *Immunodeficiency* 1994;**5**:91-112.

624 Hayoz D, Extermann M, Odermatt BF, Pugin P, Regamey C, Knecht H. Familial primary gastric lymphoma. *Gut* 1993;**34**:136-140.

625 Thorburn C, Rodriguez L, Parsonnet J. Epidemiology of gastric non-Hodgkin's lymphoma patients: parallels with Helicobacter pylori. *Helicobacter* 1996;**1**:75-78.

626 Narod S, Ford D, Devilee P, et al. Genetic heterogeneity of breast-ovarian cancer revisited. *Am J Hum Genet* 1995;**57**:957-958.

627 David KL, Steiner-Grossman P. The potential use of tumor registry data in the recognition and prevention of hereditary and familial cancer [see comments]. *N Y State J Med* 1991;**91**:150-152.

628 Bulow S, Burn J, Neale K, Northover J, Vasen H. The establishment of a polyposis register. *Int J Colorectal Dis* 1993;**8**:34-38.

629 Bussey HJ. *Familial polyposis coli*. Baltimore: The Johns Hopkins University Press, 1975;

630 Bulow S. The Danish Polyposis Register. Description of the methods of detection and evaluation of completeness. *Dis Colon Rectum* 1984;**27**:351-355.

631 Belinson JL, Okin C, Casey G, et al. The familial ovarian cancer registry: progress report. *Cleve Clin J Med* 1995;**62**:129-134.

632 Piver MS, Mettlin CJ, Tsukada Y, Nasca P, Greenwald P, McPhee ME. Familial ovarian cancer registry. *Obstet Gynecol* 1984;**64**:195-199.

633 Hodgson SV, Harocopos C, Gaglia P. Screening results in a family cancer clinic: Five years experience. *Anticancer Res* 1993;**13**:2581-2585.

634 Thompson JA, Wiesner GL, Sellers TA, et al. Genetic services for familial cancer patients: a survey of National Cancer Institute cancer centers. *J Natl Cancer Inst* 1995;**87**:1446-1455.

635 Evans DGR, Cuzick J, Howell A. Cancer genetics clinics. *Eur J Cancer* 1996;**32A**:391-392.

636 Lemon SJ, Tinley ST, Fusaro RM, Lynch HT. Cancer risk assessment in a hereditary cancer prevention clinic and its first year's experience. *Cancer* 1997;**80**:606-613.

637 Jarvinen HJ, Husa A, Aukee S, Laitinen S, Matikainen M, Havia T. Finnish registry for familial adenomatosis coli. *Scand J Gastroenterol* 1984;**19**:941-946.

638 Bulow S, Bulow C, Nielsen TF, Karlsen L, Moesgaard F. Centralized registration, prophylactic examination, and treatment results in improved prognosis in familial adenomatous polyposis. Results from the Danish Polyposis Register. *Scand J Gastroenterol* 1995;**30**:989-993.

639 Kelloff GJ, Hawk ET, Karp JE, et al. Progress in clinical chemoprevention. *Semin Oncol* 1997;**24**:241-252.

640 Iselius L, Wilking N. Screening for familial cancer. In: Weber W, Mulvihill JJ, Narod SA, eds. *Familial cancer management*. New York: CRC press, 1996;115-129.

641 Weber W. Chemoprevention in familial cancer. In: Weber W, Mulvihill JJ, Narod SA, eds. *Familial cancer management*. New York: CRC press, 1996;131-136.

642 Lerman C, Croyle RT. Emotional and behavioral responses to genetic testing for susceptibility to cancer. *Oncology* 1996;**10**:191-199.

643 Hudson KL, Rothenberg KH, Andrews LB, Kahn MJE, Collins FS. Genetic discrimination and health insurance: an urgent need to reform. *Science* 1995;**270**:391-393.

644 Culliton BJ. Genes and discrimination. *Nat Med* 1995;**5**:385

645 Lapham EV, Kozma C, Weiss JO. Genetic discrimination: perspectives of consumers. *Science* 1996;**274**:621-624.

646 Muller H. Genetic counseling and cancer. In: Weber W, Mulvihill JJ, Narod SA, eds. *Familial cancer management*. New York: CRC press, 1996;107-116.

647 Geller G, Botkin JR, Green MJ, et al. Genetic testing for susceptibility to adult-onset cancer: The process and content of informed consent. *JAMA* 1997;**277**:1467-1474.

648 Giardiello FM, Brensinger JD, Petersen GM, et al. The use and interpretation of commercial APC gene testing for familial adenomatous polyposis. *N Engl J Med* 1997;**336**:823-827.

649 Boder E. Ataxia-telangiectasia: some historic, clinical and pathologic observations. *Birth Defects* 1975;XI:255-270.

650 Swift M. Genetic aspects of ataxia-telangiectasia. *Immunodef rev* 1990;**2**:67-81.

651 Gorlin RJ, Cohen MM, Jr., Condon LM, Burke BA. Bannayan-Riley-Ruvalcaba syndrome. *Am J Med Genet* 1992;**44**:307-314.

652 Fargnoli MC, Orlow SJ, Semel-Concepcion J, Bolognia JL. Clinicopathologic findings in the Bannayan-Riley-Ruvalcaba syndrome. *Arch Dermatol* 1996;**132**:1214-1218.

653 Kimonis VE, Goldstein AM, Pastakia B, et al. Clinical manifestations in 105 persons with nevoid basal cell carcinoma syndrome. *Am J Med Genet* 1997;**69**:299-308.

654 Wiedemann HR. Tumours and hemihyperstrophy associated with Wiedemann-Beckwith syndrome. *Eur J Pediatr* 1983;**141**:129

655 Cohen MM, Jr. A comprehensive and critical assessment of overgrowth and overgrowth syndromes. *Adv Hum Genet* 1989;**18**:181-303.

656 Sotelo-Avila C, Gooch III WM. Neoplasms associated with the Beckwith-Wiedemann Syndrome. In: Rosenberg HS, ed. *Perspectives in pediatric pathology*. Chicago: Year Book Med Publ, 1973;255-272.

657 German J. Bloom's syndrome. *Dermatol Clin* 1995;**13**:7-18.

658 Stratakis CA, Courcoutsakis NA, Abati A, et al. Thyroid gland abnormalities in patients with the syndrome of spotty skin pigmentation, myxomas, endocrine overactivity, and schwannomas (Carney complex). *J Clin Endocrinol Metab* 1997;**82**:2037-2043.

659 Carney JA, Stratakis CA. Epithelioid blue nevus and psammomatous melanotic schwannoma: The unusual pigmented skin tumors of the Carney complex. *Semin Diagn Pathol* 1998;**15**:216-224.

660 Stratakis CA, Kirschner LS, Carney JA. Carney complex: Diagnosis and management of the complex of spotty skin pigmentation, myxomas, endocrine overactivity, and schwannomas. *Am J Med Genet* 1998;**80**:183-185.

661 Sander CA, Medeiros LJ, Weiss LM, Yano T, Sneller MC, Jaffe ES. Lymphoproliferative lesions in patients with common variable immunodeficiency syndrome. *Am J Surg Pathol* 1992;**16**:1170-1182.

662 Dokal I. Annotation: Dyskeratosis congenita: an inherited bone marrow failure syndrome. *Br J Hematol* 1996;**92**:775-779.

663 Hanssen AMN, Fryns JP. Cowden syndrome. *J Med Genet* 1995;**32**:117-119.

664 Sheldon CD, Hodson ME, Carpenter LM, Swerdlow AJ. A cohort study of cystic fibrosis and malignancy. *Br J Cancer* 1993;**68**:1025-1028.

665 Schoni MH, Maisonneuve P, Schoni-Affolter F, Lowenfels AB. Cancer risk in patients with cystic fibrosis: The European data. *J R Soc Med* 1996;**89**:38-43.

666 Neglia JP, Fitzsimmons SC, Maisonneuve P, et al. The risk of cancer among patients with cystic fibrosis. *N Engl J Med* 1995;**332**:494-499.

667 Pelletier J, Bruening W, Kashtan CE, et al. Germline mutations in the Wilm's tumor suppressor gene are associated with abnormal urogenital development in denys-drash syndrome. *Cell* 1991;**67**:437-447.

668 Pinheiro M, Freire-Maia N. Ectodermal dysplasias: a clinical classification and a causal review. *Am J Med Genet* 1994;**53**:153-162.

669 Schwartz RA, Birnkrant AP, Rubenstein DJ, et al. Squamous cell carcinoma in dominant type epidermolysis bullosa dystrophica. *Cancer* 1981;**47**:615-620.

670 Song IC, Dicksheet S. Management of squamous cell carcinoma in a patient with dominant-type epidermolysis bulosa dystrophica: a surgical challenge. *Plast Reconstr Surg* 1985;**75**:732-736.

671 Grubauer G, Hintner H, Klein G, Fritsch P. [Acquired, surface giant nevus cell nevi in generalized, atrophic, benign epidermolysis bullosa]. *Hautarzt* 1989;**40**:523-526.

672 Bergman W, Watson P, de Jong J, Lynch HT, Fusaro RM. Systemic cancer and the FAMMM syndrome. *Br J Cancer* 1990;**61**:932-936.

673 Nishida T, Hirota S, Taniguchi M, et al. Familial gastrointestinal stromal tumours with germline mutation of the KIT gene. *Nat Genet* 1998;**19**:323-324.

674 Jeevaratnam P, Cottier DS, Browett PJ, van de Water NS, Pokos V, Jass JR. Familial giant hyperplastic polyposis predisposing to colorectal cancer: A new hereditary bowel cancer syndrome. *J Pathol* 1996;**179**:20-25.

675 Loh KC. Familial nonmedullary thyroid carcinoma: A meta-review of case series. *Thyroid* 1997;**7**:107-113.

676 Burgess JR, Duffield A, Wilkinson SJ, et al. Two families with an autosomal dominant inheritance pattern for papillary carcinoma of the thyroid. *J Clin Endocrinol Metab* 1997;**82**:345-348.

677 Canzian F, Amati P, Harach HR, et al. A gene predisposing to familial thyroid tumors with cell oxyphilia maps to chromosome 19p13.2. *Am J Hum Genet* 1998;**63**:1743-1748.

678 Koch B. Familial fibrocystic pulmonary dysplasia: observations in one family. *Can Med Assoc J* 1965;**92**:801-808.

679 Stratton MR. Recent advances in understanding of genetic susceptibility to breast cancer. *Hum Mol Genet* 1996;**5**:1515-1519.

680 Hoskins KF, Stopfer JE, Calzone KA, et al. Assessment and counseling for women with a family history of breast cancer: A guide for clinicians. *JAMA* 1995;**273**:577-585.

681 Whitelaw SC, Murday VA, Tomlinson IPM, et al. Clinical and molecular features of the hereditary mixed polyposis syndrome. *Gastroenterology* 1997;**112**:327-334.

682 Lynch HT, Smyrk T, Lynch J. An update of HNPCC (Lynch syndrome). *Cancer Genet Cytogenet* 1997;**93**:84-99.

683 Lowenfels AB, Maisonneuve P, DiMagno EP, et al. Hereditary pancreatitis and the risk of pancreatic cancer. *J Natl Cancer Inst* 1997;**89**:442-446.

684 Moll AC, Imhof SM, Bouter LM, et al. Second primary tumors in patients with hereditary retinoblastoma: A register-based follow-up study, 1945-1994. *Int J Cancer* 1996;**67**:515-519.

685 Delaporte E, N'guyen-Mailfer C, Janin A, et al. Keratoderma with scleroatrophy of the extremities or sclerotylosis (Huriez syndrome): a reappraisal. *Br J Dermatol* 1995;**133**:409-416.

686 Inoue H, Miki H, Oshimo K, et al. Familial hyperparathyroidism associated with jaw fibroma: Case report and literature review. *Clin Endocrinol* 1995;**43**:225-229.

687 Cunningham-Rundles C, Pudifin DJ, Armstrong D, Good RA. Selective IgA deficiency and neoplasia. *Vox Sang* 1980;**38**:61-67.

688 Gatti RA, Good RA. Occurrence of malignancy in immunodeficiency diseases. A literature review. *Cancer* 1971;**28**:89-98.

689 Cohen PR, Kohn SR, Kurzrock R. Association of sebaceous gland tumors and internal malignancy: the Muir-Torre syndrome. *Am J Med* 1991;**90**:606-613.

690 Shepherd JJ. The natural history of multiple endocrine neoplasia type 1: highly uncommon or highly unrecognized? *Arch Surg* 1991;**126**:935-952.

691 Chandrasekharappa SC, Guru SC, Manickam P, et al. Positional cloning of the gene for multiple endocrine neoplasia-type 1. *Science* 1997;**276**:404-407.

692 Eng C. The RET proto-oncogene in multiple endocrine neoplasia type 2 and Hirschsprung's disease. *N Engl J Med* 1996;**335**:943-951.

693 Morrison PJ, Nevin NC. Multiple endocrine neoplasia type 2B (Mucosal neuroma syndrome, Wagenmann-Froboese syndrome). *J Med Genet* 1996;**33**:779-782.

694 Chakrabarty KH, Perks AG. Ferguson-Smith syndrome: the importance of long term follow-up. *Br J Plast Surg* 1996;**49**:497-498.

695 Blatt J, Jaffe R, Deutsch M, Adkins JC. Neurofibromatosis and childhood tumors. *Cancer* 1986;**57**:1225-1229.

696 Poyhonen M, Niemela S, Herva R. Risk of malignancy and death in neurofibromatosis. *Arch Pathol Lab Med* 1997;**121**:139-143.

697 Pollack IF, Mulvihill JJ. Neurofibromatosis 1 and 2. *Brain Pathol* 1997;**7**:823-836.

698 Mulvihill JJ. Malignancy: epidemiologically associated cancers. In: Huson SM, Hughes RAC, eds. *The Neurofibromatoses: a pathogenetic and clinical overview*. London: Chapman & Hall, 1994;305-315.

699 Friedman JM, Birch PH. Type 1 neurofibromatosis: A descriptive analysis of the disorder in 1,728 patients. *Am J Med Genet* 1997;**70**:138-143.

700 Konishi F, Wyse NE, Muto T, et al. Peutz-Jeghers polyposis associated with carcinoma of the digestive organs: report of three cases and review of the literature. *Dis Colon Rectum* 1987;**30**:790-799.

701 Stevens HP, Kelsell DP, Leigh IM, Ostlere LS, MacDermot KD, Rustin MHA. Punctate palmoplantar keratoderma and malignancy in a four-generation family. *Br J Dermatol* 1996;**134**:720-726.

702 Bennion SD, Patterson JW. Keratosis punctata palmaris et plantaris and adenocarcinoma of the colon: a possible familial association of punctate keratoderma and gastrointestinal malignancy. *J Am Acad Dermatol* 1984;**10**:587-591.

703 Leonard A, Craft AW, Moss C, Malcolm AJ. Osteogenic sarcoma in the Rothmund-Thomson syndrome. *Med Pediatr Oncol* 1996;**26**:249-253.

704 Garganta CL, Bodurtha JN. Report of another family with simpson-golabi-behmel syndrome and a review of the literature. *Am J Med Genet* 1992;**44**:129-135.

705 Pampiglione G, Moynahan EJ. The tuberous sclerosis syndrome: clinical and EEG studies in 100 children. *J Neurol Neurosurg Psychiatry* 1976;**39**:666-673.

706 Hamilton SR, Liu B, Parsons RE, et al. The molecular basis of Turcot's syndrome. *N Engl J Med* 1995;**332**:839-847.

707 Itoh H, Ohsato K, Yao T, Iida M, Watanabe H. Turcot's syndrome and its mode of inheritance. *Gut* 1979;**20**:414-419.

708 Neumann HPH, Lips CJ, Hsia YE, Zbar B. Von Hippel-Lindau syndrome. *Brain Pathol* 1995;**5**:181-193.

709 Ry Andersen S, Geertinger P, Larsen HW, et al. Aniridia, cataract and gonadoblastoma in a mentally retarded girl with deletion of chromosome 11: a clinicopathological case report. *Ophthalmologica* 1978;**176**:171-177.

710 Epstein CJ, Martin GM, Schultz AL, Motulsky AG. Werner's syndrome: a review of its symptomatology,natural,history,pathologic features,genetics and relationship to the natural aging process. *Medicine* 1966;**45**:177-221.

711 Lambert WC, Lambert MW. Disorders of DNA and chromosomal instability. In: Harper J, ed. *Inherited skin disorders: The genodermatoses.* Oxford: Reed Educational & Professional Publishing ltd. 1996;276-283.

712 van der Meer JW, Weening RS, Schellekens PT, van Munster IP, Nagengast FM. Colorectal cancer in patients with X-linked agammaglobulinaemia. *Lancet* 1993;**341**:1439-1440.

713 van der Meer JW, Zegers BJ. Agammaglobulinaemia. *Neth J Med* 1994;**45**:250-256.

714 Seemayer TA, Grierson H, Pirruccello SI, et al. X-linked lymphoproliferative disease. *Am J Dis Child* 1993;**147**:1242-1245.

715 Brownstein MH, Wolf M, Bikowski JB. Cowden's disease: a cutaneous marker of breast cancer. *Cancer* 1978;**41**:2393-2398.

716 Walton BJ, Morain WD, Baughman RD, Jordan A, Crichlow RW. Cowden's disease: a further indication for prophylactic mastectomy. *Surgery* 1986;**99**:82-86.

717 Trau H, Schewach-Millet M, Fisher BK, Tsur H. Peutz-Jeghers syndrome and bilateral breast carcinoma. *Cancer* 1982;**50**:788-792.

718 Lobaccaro JM, Lumbroso S, Belon C, et al. Male breast cancer and the androgen receptor gene. *Nat Genet* 1993;**5**:109-110.

719 Wooster R, Mangion J, Eeles R, et al. A germline mutation in the androgen receptor gene in two brothers with breast cancer and Reifenstein syndrome. *Nat Genet* 1992;**2**:132-134.

720 Risinger JI, Barrett JC, Watson P, Lynch HT, Boyd J. Molecular genetic evidence of the occurrence of breast cancer as an integral tumor in patients with the hereditary nonpolyposis colorectal carcinoma syndrome. *Cancer* 1996;**77**:1836-1843.

721 Itoh H, Houlston RS, Harocopos C, Slack J. Risk of cancer death in first-degree relatives of patients with hereditary non-polyposis cancer syndrome (Lynch type II): a study of 130 kindreds in the United Kingdom. *Br J Surg* 1990;**77**:1367-1370.

722 Lashner BA, Riddell RH, Winans CS. Ganglioneuromatosis of the colon and extensive glycogenic acanthosis in Cowden's disease. *Dig Dis Sci* 1986;**31**:213-216.

723 Marra G, Armelao F, Vecchio FM, Percesepe A, Anti M. Cowden's disease with extensive gastrointestinal polyposis. *J Clin Gastroenterol* 1994;**18**:42-47.

724 Lynch HT, Smyrk T, Mcginn T, et al. Attenuated familial adenomatous polyposis (AFAP): A phenotypically and genotypically distinctive variant of FAP. *Cancer* 1995;**76**:2427-2433.

725 Giardiello FM, Hamilton SR. Hereditary mixed polyposis syndrome: A zebra or a horse dressed in pinstripes. *Gastroenterology* 1997;**112**:643-645.

726 Thomas HJW, Whitelaw SC, Cottrell SE, et al. Genetic mapping of the hereditary mixed polyposis syndrome to chromosome 6q. *Am J Hum Genet* 1996;**58**:770-776.

727 Tops CMJ, Vasen HFA, van Berge Henegouwen G, et al. Genetic evidence that Turcot syndrome is not allelic to familial adenomatous polyposis. *Am J Med Genet* 1992;**43**:888-893.

728 Lynch HT, Smyrk TC, Watson P, et al. Hereditary flat adenoma syndrome: a variant of familial adenomatous polyposis. *Dis Colon Rectum* 1992;**35**:411-421.

729 Eeles RA. Germline mutations in the TP53 gene. *Cancer Surv* 1995;**25**:101-124.

730 Schwartz RA. Basal-cell-nevus syndrome and gastrointestinal polyposis. *N Engl J Med* 1978;**299**:49

731 O'Riordain DS, O'Brien T, Crotty TB, Gharib H, Grant CS, van Heerden JA. Multiple endocrine neoplasia type 2B: More than an endocrine disorder. *Surgery* 1995;**118**:936-942.

732 Grouls V, Vaih S, Bindewald H. Type 1 neurofibromatosis of the colon in von Recklinghausen's disease. *Pathologe* 1996;**17**:133-138.

733 Devroede G, Lemieux B, Masse S, Lamarche J, Herman PS. Colonic hamartomas in tuberous sclerosis. *Gastroenterology* 1988;**94**:182-188.

734 Gould SR, Stewart JB, Temple LN. Rectal polyposis in tuberous sclerosis. *J Ment Def Res* 1990;**34**:465-473.

735 Foster MA, Kilcoyne RF. Ruvalcaba-Myhre-Smith syndrome: a new consideration in the differential diagnosis of intestinal polyposis. *Gastrointest Radiol* 1986;**11**:349-350.

736 Marsh DJ, Dahia PLM, Zheng ZM, et al. Germline mutations in PTEN are present in Bannayan-Zonana syndrome. *Nat Genet* 1997;**16**:333-334.

737 Laken SJ, Petersen GM, Gruber SB, et al. Familial colorectal cancer in Ashkenazim due to a hypermutable tract in APC. *Nat Genet* 1997;**17**:79-83.

738 Woodage T, King SM, Wacholder S, et al. The APC 11307K allele and cancer risk in a community-based study of Ashkenazi Jews. *Nat Genet* 1998;**20**:62-65.

739 Frayling IM, Beck NE, Ilyas M, et al. The APC variants I1307K and E1317Q are assocaited with colorectal tumors, but not always with a family history. *Proc Natl Acad Sci U S A* 1998;**95**:10711-10727.

740 Wacholder S, Hartge P, Struewing JP, et al. The kin-cohort study for estimating penetrance. *Am J Epidemiol* 1998;**148**:623-630.

741 Cohen PR, Kohn SR, Davis DA, Kurzrock R. Muir-Torre syndrome. *Dermatol Clin* 1995;**13**:79-89.

742 Boland CR, Thibodeau SN, Hamilton SR, et al. National Cancer Institute Workshop on Microsatellite Instability for Cancer Detection and Familial Predisposition: Development of International Criteria for the Determination of Microsatellite Instability in Colorectal Cancer. Cancer Res., 58 : 5248-5257, 1998.

743 Cravo ML, Fidalgo PO, Lage PA, et al. Validation and simplification of Bethesda guidelines for identifying apparently sporadic forms of colorectal carcinoma with microsatellite instability. *Cancer* 1999;**85**:779-785.

744 Winawer SJ, Fletcher RH, Miller L, et al. Colorectal cancer screening: clinical guidelines and rationale. *Gastroenterology* 1997;**112**:594-642.

745 Hodgson SV, Bishop DT, Dunlop MG, Evans DG, Northover JM. Suggested screening guidelines for familial colorectal cancer. *J Med Screen* 1995;**2**:45-51.

746 Winawer SJ, Stjohn DJ, Bond JH, et al. Prevention of colorectal cancer: Guidelines based on new data. *Bull Who* 1995;**73**:7-10.

747 Winawer SJ, Fletcher RH, Miller L, et al. Colorectal cancer screening: Clinical guidelines and rationale (Vol 112, pg 594, 1997): corrections. *Gastroenterology* 1997;**112**:1060

748 Burke W, Petersen G, Lynch P, et al. Recommendations for follow-up care of individuals with an inherited predisposition to cancer: 1. Hereditary nonpolyposis colon cancer. *JAMA* 1997;**277**:915-919.

749 Markowitz AJ, Winawer SJ. Screening and surveillance for colorectal carcinoma. *Hematol Oncol Clin N Amer* 1997;**11**:579

Chapter 3
Accuracy of the family history of cancer: clinical implications.

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3.1 Abstract

Background: The family history is the cornerstone of clinical genetic diagnosis and management in cases of familial cancer. The soundness of medical decision making can be compromised if family reports on affected relatives are inaccurate. Although it is very time consuming, family histories are therefore routinely verified.

Aim: We performed a retrospective study to assess the accuracy of the family history as collected in our family cancer clinic and investigated whether verification through medical records had an additional value with regard to clinical genetic management.

Design: Files of a consecutive series of 120 families, referred because of familial/hereditary cancer to our clinic between 1977 and 1997, were used for analysis. The accuracy of the family history with respect to cancer type and age at diagnosis was assessed for 383 tumours. Moreover, we investigated a range of medical and sociodemografic parameters for their possible influence on this accuracy. Finally, the impact of the verification efforts on the clinical genetic diagnosis and management was evaluated according to tumour type and degree of kinship.

Results: The accuracy according to cancer type showed marked variation, ranging from 93% and 89% for breast cancer and colorectal cancer, respectively, to 42% and 37% for extra-colorectal alimentary tract cancer and uterine cancer. The accuracy was related to the degree of kinship of the affected relative, but not to age and gender of the consultand, nor to reason for referral or a personal history of cancer. Age at diagnosis was reported accurately in 97% of cases, and multiple primary tumours were correctly reported in 94% of cases. In 6 out of 120 families verification data changed clinical genetic management, in 5 of these the genetic risk became reduced.

Conclusion: Although verification of all reported cancer cases in a family remains the 'golden standard' for clinical as well as research purposes, efforts to verify the frequent reports on breast cancer can be limited without seriously compromising medical decision making. This seems currently not to be the case for other types of cancer.

3.2 Introduction

There is a growing demand for clinical genetic analysis of families suspected of having hereditary cancer. The family history of cancer plays a central role in confirming or rejecting this suspicion. Unfortunately, details of a family history of cancer are not necessarily accurate and may lead to wrong clinical decisions and counselling, if they are left unverified. However, verification is time-consuming and a rapidly growing burden to the clinic. Therefore, it would be helpful if verification could be minimised without compromising the soundness of clinical genetic decision making. If we would know the accuracy of reported tumour types, ages at tumour diagnosis, and presence of multiple primary tumours, all important for the genetic interpretation of a family history of cancer, and if we also knew the overall impact of verifications on that interpretation, then this might help in differentiating the clinical need for verification. Also, it would help in estimating the accuracy of family reports in those cases, where routine destruction of older medical records makes verification impossible.

Only a small number of studies have addressed the accuracy of the family history of cancer and mainly when it was collected for epidemiological purposes [1-11]. Moreover, these studies verified the history of only the first- and second-degree relatives and usually did not allow the person interviewed to consult their relatives. In the setting of a family cancer clinic the opposite is true: there is no restriction with regard to degrees of kinship and extensive time is given to check family history with relatives. Studies on the accuracy of the family history of cancer as obtained by methods commonly used in family cancer clinics are rare [12,13].

In this retrospective study we investigated the accuracy of the family history of cancer, taken in the setting of the familial cancer clinic in Groningen.

3.3 Material and Methods

3.3.1 General procedure of taking and verifying the family history.

Each consultand (the name introduced to refer to individuals seeking genetic testing and counselling [14,15], as opposed to consultant) referred to the Department of Medical Genetics in Groningen, The Netherlands, because of a family history of cancer is sent a form at least 4 weeks before the first visit to the Department. On that form, the consultand is asked to write down family details (number and type of

all first, second and most third degree relatives; names, dates of birth and death of first and second-degree relatives; types of cancer and ages at tumour diagnosis in all relatives). A leaflet accompanies the form and encourages the consultand to supply detailed information on the cases of cancer in the family, excluding metastases. The form is returned before the first visit and a pedigree is drawn based on the given information.

During the first visit, details of the family history are discussed by the consultand and a clinical geneticist, who explains (again) to the consultand the difference between primary tumours and metastases. As a result, a number of tumours may be reclassified during that visit as (probable) metastases (predominantly in the liver, skeleton, lungs and brain).

Details of the family history are subsequently verified. For that purpose, a written consent has to be obtained from each living relative whose information has to be verified. Regarding deceased relatives, in the Netherlands the physician of those relatives decides whether or not to supply the information. The sources for verification are the patient's medical records, preferably and mainly the pathology reports, from the hospitals and general practitioners.

In the Netherlands, patient records are stored for a legal minimum of 10 years after the last clinical contact, and may be destroyed after that period. Academic hospitals have a general policy of keeping medical records until 115 years after the patient's date of birth. In our country, only the physicians who originally filled out the death certificates have legal access to them for clinical purposes (other rules apply to epidemiological studies). As it is virtually impossible to identify and contact these physicians years after filing the certificates, it makes death certificates useless for verification purposes. For practical reasons, negative histories of cancer in relatives are almost never verified.

3.3.2 Retrospective family history verification study:

A consecutive series of 139 consultands from 129 families referred to the Department between 1977 and 1997 because of a family history of cancer was included in this study. Excluded were consultands whose family histories had already been verified at other institutes. If a family was already included in the study, new consultands from this family were excluded unless they were only distantly related to the consultands already included and had collected details on their family history separately from the other consultands by contacting different relatives for that purpose (16 consultands from 6 families fell within this category).

Data on the following items were retrospectively collected from the Department's records of these consultands and their families and stored in a computer database (dBase IV, © Borland): (a) family identification no.; (b) the reported primary tumour types in the consultands and their relatives;(c) the type (*e.g.* parent) and degree of kinship with those relatives (*e.g.* 1st); (d) the reported ages at tumour diagnosis (categories with 5-year intervals); (e) the clinical diagnosis if the family history had been believed 'at face value' (and before possible additional physical examination/laboratory testing); (f) the outcome of verification of the reported tumours (type, age at diagnosis); (g) the reasons for non-verification; and (h) the clinical diagnosis directly after verification (and before possible additional physical examination/laboratory testing). In addition, the following items were included because they were considered potential confounders: (I) gender of the consultand; (II) age of the consultand; (III) whether or not the consultand had a personal history of cancer and if so what type; (IV) the reason for referral; and (V) years past after diagnosis for each of the reported tumours.

The family history for a particular cancer was considered accurate if both organ and disease type (*i.e.* cancer) were correct. If the history had been reported as a wider category (*e.g.* 'leukaemia', 'lymphoma', 'abdominal cancer' or 'uterine cancer') rather than as a more specific diagnosis (*e.g.* CML, NHL, pancreatic cancer and cervical or endometrial cancer), then the report was considered accurate if the confirmed diagnosis fell within that wider category. Reported age at diagnosis was considered true if it differed less than 5 years from the true age.

The chi-square (χ^2) test for heterogeneity was used to test for statistical differences in accuracy between the kinship-tumour type classes for each of the above mentioned parameters.

In case sample sizes were too small for the chi-square test (breast, ovarian and colorectal cancer groups), the Fischer's exact test was used instead. The chi-square test for trends was used to look for trends of increasing or decreasing accuracy with increasing or decreasing degree of kinship. P-values below 0.05 were considered statistically significant. All tests were performed using the S.P.S.S. computer program (version 6.1, © Statistical Package for the Social Sciences,Inc.).

3.4 Results

A total of 886 tumours had been reported. Of these reported tumours, 383 (43%) from a total of 120 families had been verified. The results for these verified cases are shown in table 1. Although we had recorded the specific type of relative within each degree of kinship (*e.g.* siblings, parents and children within the 1st-degree category), the sample sizes within each of these subcategories were too small to be used for statistical analysis. We therefore only present the data on the degree of kinship. Numbers of reported tumours within the category 'other' in table 1 were also relatively small. Therefore, differences of accuracy between different degrees of kinship were not calculated for the individual tumour types in this category.

When all degrees of kinship were pooled, significant heterogeneity ($P<0.0001$) was observed for the accuracy of the reported tumour types/subcategories: accuracy for breast cancer was 93%, for colorectal cancer 89%, for central nervous system cancer 88%, for respiratory tract cancer 78%, for urogenital tract cancer 78%, for ovarian cancer 71%, for haematolymphoid malignancies 64%, for extra-colorectal alimentary tract cancer 42%, for uterine cancer (including uterine cancer-not further specified, endometrial cancer and cervical cancer) 37% and for the rest group 63%.

The accuracy of reported cases was not related to the gender of the consultand ($P=0.59$), age of the consultand ($P=0.34$), a personal history of cancer ($P=0.62$), the time past after tumour diagnosis ($P=0.1$) and early age at diagnosis of the tumour ($P=0.50$). If the reason for referral was suspicion of hereditary breast-ovarian cancer, then accuracy of reports on breast ($P=0.72$) and ovarian cancer ($P=1.0$) were not significantly different from those in families which had been referred for other tumour syndromes.

Table 1 Accuracy of the family history depending on degree of kinship and tumour type

	degree of kinship						
	0	1st	2nd	3rd	4th	All degrees	P value for heterog. \ trend
Type of cancer:	% accurate	(of n reported	tumours)				
Breast	100 (15)	94 (69)	90 (31)	75 (8)	100 (2)	93 (125)	0.25 \ n.a.
Colorectal	100 (14)	91 (33)	100 (15)	57 (7)	60 (5)	89 (74)	0.006 \ n.a.
Ovarian	100 (5)	67 (15)	60 (5)	50 (2)	100 (1)	71 (28)	0.52 \ n.a.
Other	85 (26)	70 (63)	52 (42)	44 (9)	38 (16)	63 (156)	0.006 \<0.0001
All types	95 (60)	91 (180)	79 (93)	68 (26)	60 (24)	78 (383)	<0.0001\<0.0001

Degree of kinship: 0 = the consultands themselves.; 1 = children, siblings and parents; 2 = grandchildren, grandparents, nephews, nieces, aunts and uncles, 3= sibling's grandchildren, cousins, great-grandparents and grandparent's siblings.
For details on the category cancer type 'other', consult the results section.
Accurate = reported disease type (malignant) and organ type both confirmed.
Fischer's exact test was used to test for differences (heterog. = heterogeneity) in the categories breast cancer, colorectal cancer and ovarian cancer, because of smaller sample sizes. In these categories testing for trends was not applicable (n.a.)
The categories 'other tumours' and 'all tumour types' were tested using the chi-square method.

If the reason for referral was suspicion of hereditary non-polyposis colorectal cancer, then accuracy of reports on colorectal (P=0.36) and uterine cancer (P=0.80) were not significantly different from those in families which had been referred for other tumour syndromes.

In 72% of the reported tumours an age at diagnosis was given. This depended significantly on the degree of kinship: ages were reported more often in closer relatives (P<0.001). Of these reported ages, 97% were correct (dependence on degree of kinship could not be calculated due to the small number of incorrectly reported ages). Of the 32 reports on multiple primary tumours, the fact that they were indeed multiple was accurate in 94% of cases (dependence on degree of kinship could not be calculated due to the small number of these reports).

Differences were observed between the clinical diagnosis (solely based on family history) before and after verification in 17 out of 120 families (14%), listed in table 2. In six of those families (c, g (3x), h and i), *i.e.* 5% of the total number of families, this difference led to a change in indications for DNA testing, counselling and follow-up. In 5 out of 6 cases it meant a decrease of genetic risk, in 1 case (family i) it resulted in an increase).

Non-verified cases included 503 tumours or 57% of the total number of reported tumours. No effort to obtain written consent and to subsequently try to verify cases was made in 71% of these cases because (a) it was certain that the files had been destroyed; (b) consultands did not have any contact with the relatives concerned, so that no consent could be obtained; (c) the reports concerned relatives from a branch of the family not of interest to the genetic investigation; or (d) the reported cases were late-onset common type tumours in distant relatives not likely to be of interest with regard to the reason for referral. Written consent was obtained, but no information could be retrieved in 16% of the non-verified cases. Written consent was asked for, but not given in 9% of the non-verified cases. In the remaining 4% of cases the reason for non-verification could not be recollected from our records.

Table 2 Families with differences between diagnoses before and after verification of the family history

Diagnosis before verification	Diagnosis after verification	number
a) hereditary breast-ovarian cancer (HBOC)	probably* HBOC	4
b) suggestive** of HBOC	HBOC	2
c) suggestive** of HBOC	not suggestive of any inherited cancer predisposition	1
d) hereditary breast cancer (HBC)	probably* HBC	2
e) HBC	suggestive** of HBC	1
f) hereditary non-polyposis colorectal cancer (HNPCC)	probably* HNPCC	2
g) familial clustering of cancer	not suggestive of any inherited cancer predisposition	3
h) familial clustering of acute lymphocytic leukaemia (ALL)	non-familial ALL	1
i) possibly basal cell nevus syndrome***	basal cell nevus syndrome	1

Each diagnosis shown is based solely on family history (either unverified or verified), which is the basis for further medical decisions on e.g. DNA testing and follow-up. Diagnosis before verification reflects diagnosis if family history would have been believed to be completely accurate.
Differences between diagnosis before and after verification were observed in 17 out of 120 families. These differences were clinically important with regard to medical decision making in only 6 families (c, g (3x), h and i)
** probably HB(O)C/HNPCC refers to the post-verification situation where the clinical diagnosis HB(O)C or HNPCC would have been made if we assumed the family history to be accurate, however information could not be collected on all relevant cases in the family (required for formal diagnosis).*
*** suggestive of HBC refers to the situation that although formal criteria for HBC have not been met (at least 3 verified breast cancer cases in 2 successive generations, 1 of the affected relatives being a first-degree relative of the other 2 (second-degree relative counts as first-degree in case of transmission through the paternal line), several facts point towards the HBC diagnosis (e.g. multiple early-onset primary breast cancer).*
**** possibly basal cell nevus syndrome refers to the fact that family history was suggestive, but not detailed enough to make the diagnosis.*

3.5 Discussion

Our data show that the accuracy of a family history of cancer, taken in the setting of a family cancer clinic, depends primarily on tumour type and degree of kinship. Although the accuracy of reports generally declined with increasing genetic distance between consultand and affected relative, this effect did not reach statistical significance for breast and ovarian cancer. Observations on accuracy by other authors are listed in table 3. Although study designs differ, most studies observe similar relations between accuracy and tumour type or degree of kinship. The low accuracy of reports of uterine cancer we observed, important because of its relevance to hereditary non-polyposis colorectal cancer diagnosis, confirms earlier observations of 25%-40% accuracy rates [11,16]. One explanation for these low scores may be the fact that a history of hysterectomy is interpreted by patient's relatives as being an 'operation for cancer', while in many cases that operation was indicated for other reasons (*e. g.* uterine myomas and prolapse). A similar phenomenon may account for part of the false positive reports of other tumour types as well (*e.g.* operations for benign breast disease, ovarian cysts, benign colorectal polyps and colitis ulcerosa to mention typical examples).

We had speculated that the accuracy of tumour reports could also have been influenced by several other factors:

(1) gender of the consultand, because of the observation in daily practice that women often appear to have a better recollection of family details than men. Indeed, a slightly higher accuracy of family histories taken from women has been reported [16], although the opposite has also been observed [17]; (2) age of the consultand. Higher accuracy in younger subjects has been reported [16,18]; (3) whether or not the consultand had a personal history of cancer, because it either could have made him or her more sensitive to the fact that cancer had indeed occurred in relatives [18], or could have biased the interpretation of family case histories (malignant instead of benign disease) [8,19]; (4) years past after tumour diagnosis, because recollection of events from the distant past may be difficult; (5) age at tumour diagnosis, because early-onset cancer might have had a more 'dramatic' impact on families and therefore also on recollection.; (6) reason for referral, because possibly tumours central to the reason for the referral (*e.g.* cases of breast cancer in a family referred because of a suspicion of hereditary breast cancer) are more accurately reported than others.

None of these factors was of significant influence on the accuracy of the reports on cancer in our study. The reason that factors 1-4 did not significantly influence accuracy might be that instead of a family history recollected and subsequently reported by a single person (the consultand), the family history collected in the setting of a family cancer clinic is in fact the product of the recollection and reconstruction by the consultand as well as his or her relatives.

With respect to the age at diagnosis, families often tell that the diagnosis of cancer at an early age was a great shock to them. A possible explanation for its apparent absence of influence on accuracy may be that its effect is 'levelled out' by the cumulative events in the family (early-onset as well as late-onset cancer cases), finally leading to the suspicion of hereditary cancer. Events might therefore be remembered equally.

With respect to the reason for referral, one might expect that the tumour(s) central to the reason for referral might be subject to recall bias. We did not observe this. A possible explanation is that, in our experience, families often thought that the concept of hereditary cancer implied an inherited susceptibility to *all* types of cancer. Therefore, all types were considered important by them with respect to heredity. Also, and perhaps more importantly, the fact that we motivated them to supply information on all types of cancer and gave them time to consult within their family may have helped in this respect [20].

In addition to the accuracy of reported tumour types, we looked at the age at diagnosis and at occurrence of multiple primary tumours, because these are genetically important for the interpretation of a family history of cancer. If the age at diagnosis was reported (72% of the reported tumours), it turned out to be very reliable (97%), which is in agreement with the high score (89%) observed by others [8]. Reports of multiple primary tumours were generally also accurate (94%).

In summary, family histories of colorectal and breast cancer, as well as the age at diagnosis and the occurrence of multiple primary tumours, taken in a clinical genetic setting appeared to be accurate in more than 89% of cases. Most referrals to our clinic are made because of a suspicion of hereditary breast cancer or hereditary non-polyposis colorectal cancer. Therefore, it did not come as a surprise that the impact of verification on medical management turned out to be small. The reason for this is that those tumours in a family with breast or colorectal cancer that may have been inaccurately reported, generally do not outweigh the number of accurate reports from that same family.

Because of its design, our study has two obvious limitations. First, there is the possibility that the non-verified cases in our study do not have the same accuracy as the verified ones and that the accuracy rates we found may therefore not apply to all reported tumours in a family. This might mislead physicians in the weighing of family reports in those cases where verification fails because of the absence of consent or, more importantly, because of the routine destruction of older medical records. We have, however, little reason to assume that accuracy in our non-verified cases is very different, because of the similarities between our findings and those observed in other studies which verified reports without the particular restriction of our study (*i.e.* our need to obtain written consent from relatives and the blocked access to death certificates).

The second limitation of our study is that we did not assess the accuracy of negative tumour reports. However, others have reported an accuracy of negative reports of 99% in their study on cancer in first-degree relatives [21]. We therefore expect little underreporting of cancer in families referred for their family history of cancer, that we expect to be alert to the occurrence of cancer. This may be especially true for close relatives, who are most important for the genetic evaluation.

What are the practical implications of the results from our study? Based on our findings and because of the need to increase efficiency, we have now significantly decreased the verification of reported breast cancer cases. Since referrals because of familial breast cancer are very frequent, this had a major impact on our overall verification effort. We do try, however, to verify the reports of breast cancer in our consultands, because in our opinion documentation of our own patients should be complete before we provide counselling specifically aimed at these patients. The effort to do this is relatively small, as most of this information is provided upon referral to our clinic. For basically the same reason, we also verify data on all relatives with breast cancer who participate in diagnostic DNA analysis. As a rule, breast cancer cases with a remarkable clinical presentation (*e.g.* early-age at diagnosis, multiple primary tumours) are verified because they strongly influence risk calculation [22], interpretation of diagnostic DNA test results and planning of medical management. Using this system, the chance that additional verification will lead to changes in medical management is likely to be much lower than the 5 % we found if no verification had been done at all (or the 11 % reported by Douglas *et al*[13])

One potential pitfall in our system would be the interpretation of a factitious breast cancer family history. Presumably, however, such histories are very rare [23,24]. In general, in all cases where one has reason to suspect less than usual accuracy, *e.g.* when the consultand did not have direct contact with any of the cancer patients in the family, or because of a consultand's particular psychiatric history, extra attention should be given to verification.

As a rule, we verify reported tumours other than breast cancer. We usually exclude cases, especially common types of late-onset cancer in distant relatives, if the outcome of verification is expected to contribute little to the genetic diagnosis. Typical examples are histories of late-onset lung cancer in heavy smokers, late-onset non-melanoma skin cancer of sun-exposed areas, or additional cases of colorectal cancer in a family where verified cases already allowed for the diagnosis HNPCC. Although family reports on colorectal tumours are generally accurate, we often verify these reports, especially in cases of ambiguous genetic diagnosis, because details usually unreported by the family, *e.g.* the location of the tumour (proximal?), history of benign precursor lesions (early onset adenomas?) and the specific histology (mucinous with heavy lymphocyte infiltration?) may provide information which helps to select tumour material as well as patients for DNA analysis. Some family histories are even relatively useless without collecting the histology reports. Early-onset intestinal polyps (adenomatous or hamartous?), thyroid cancer (medullary or non-medullary?), renal cancer (clear cell or non-clear cell type of renal cell cancer? transitional cell cancer of the pyelum?) are examples where, depending on the specific tumour histology, the clinical genetic differential diagnosis may change radically.

Limiting verification carries a risk that information is missed, which might have a major impact on the clinical genetic diagnosis and medical family management. We think that using our system of collecting and verifying the family history, this risk is small. Another drawback of limiting verification is that the value of pedigree information for research purposes may also be limited. If there are no organisational needs to limit verification, then we recommend verifying the complete family history (unless psychological aspects in a particular family would make this undesirable). We expect, however, that the growing options for DNA analysis will ultimately considerably weaken the need for full family history verification in the clinical setting, as diagnosing hereditary disorders through cheap and fast DNA analysis in a limited number of affected relatives will substitute thorough verification and interpretation of extended family histories.

Table 3 **Accuracy of family histories of cancer: review of published data**

all tumours combined			breast	colorectal	ovarian	comment and references
degree of kinship						
1st	2nd	3rd	all	all	all	
accuracy: percentage of correct information out of total cases (number between brackets)						
91 (180)	79 (93)	68 (26)	93 (125)	89 (74)	71 (28)	This study
84 (216)	71 (174)	71 (73)	91 (157)	88 (79)	-	Families referred to family cancer clinic, FH taken by questionnaire, verified by MR[12].
86 (n.r.)	78 (n.r.)	59 (n.r.)	95 (n.r.)			Families referred to family cancer clinic, FH taken by questionnaire, verified by MR/DC/CR[13].
78 (32)**	70 (54)**	-	100 (5)	100 (5)	100 (2)	Case(ES) study, FH taken by mailed questionnaire + PI, verified mainly by DC[9,25].
85 (252)	67 (165)	-	98 (59)	87 (47)	57 (7)	Case(GLI) study, FH taken by PI (multiple relatives), verified by MR+DC[11].
-	-	-	57 (51)	52 (46)	27 (11)	Case(COL)-control study, FH taken by PI, confirmed by GDB/CR [10,16].
n.r. (230)	-	-	>88 (n.r.)	>88 (n.r.)	-	Case(CBT)-control study, FH taken by PI, verification method n.r.[5]

	88 (2090)***	-	93 (349)	-	-	Case(LW) study, FH taken by PI (proband+ parent), verified by MR+DC[1]
	88 (100)***	-	-	-	-	Case(Mix)-control study, FH taken by mailed questionnaire, verified by MR+DC[3]
-	-	-	92 (97)	-	-	Case(BR)-control study, FH taken by PI, verified by MR[8]
88 (98)	72 (76)	-	-	-	-	Case(S) study, FH taken by PI, verified by MR+DC[6].
85-86* (n.r.)	20-36* (n.r.)	-	-	-	-	Case(TC)-control study, FH taken by unknown method, verified by DC[2].
96 (1171)	87 (553)	-	-	-	-	Case(OV)-control study, FH taken by PI, verified by CR[4].
84 (39)****	-	-	-	-	-	Case(CHN) study, FH taken by PI, verified by MR[7].

correct information = reported disease type (malignant) and organ type both confirmed.
n.r.= not reported
The studies a-c, determined accuracy of breast, colorectal and ovarian cancer for pooled first- and second- and third-degree relatives, whereas the others listed did not include third-degree relatives. This fact alone could result in lower rates found in studies a-c, compared with the others. In our study, accuracy for the three types of tumours in pooled first- and second-degree relatives only, were 93 % for breast cancer, 94 % for colorectal cancer and 65 % for ovarian cancer. We could not calculate this for the studies b and c, because the data needed to do this were unavailable.
**= 1st-degree relatives limited to parents; 2nd-degree to grand-parents*
***= 1st-degree relative or spouse*
****= mixed group of 1st- and 2nd-degree relatives*
*****= 1st-degree relatives limited to sibs*
Comments: FH= family history; PI= personal interview; MR= medical records, DC= death certificates, CR= cancer registry, GDB/CR= genealogic database linked to CR. Codes referring to the type of proband in case(-control) studies: BR= breast cancer; CBT= childhood brain tumour; CHN= childhood head & neck condition treated with irradiation; ES= Ewing sarcoma; GLI= glioma; LW= lactating women; Mix= patients with a wide range of malignancies; OV= ovarian cancer; S= soft-tissue and bone sarcoma.

Not listed in this table is the study of 1040 reported melanomas in first-degree relatives of melanoma patients by Aitken et al.[18], who took the family history using mailed questionnaires and observed only 60% accuracy.

Each family cancer clinic will have to decide on their own protocols for taking family histories and verifying them, depending on local resources, clinical and organisational needs, legislation and research interests. We hope that our findings may be of use in that respect.

3.6 References

1 Albert S, Child M. Familial cancer in the general population. *Cancer* 1977;**40**:1674-1679.

2 Harris R, Linn MW, Hunter K. How valid are patients reports of family history of illness? *Meth inform med* 1980;**19**:162-164.

3 Schneider NR, Chaganti SR, German J, Chaganti RS. Familial predisposition to cancer and age at onset of disease in randomly selected cancer patients. *Am J Hum Genet* 1983;**35**:454-467.

4 Koch M, Gaedke H, Jenkins H. Family history of ovarian cancer patients: a case-control study. *Int J Epidemiol* 1989;**18**:782-785.

5 Kuijten RR, Bunin GR, Meadows T. Validation and accuracy of cancer reporting in first-degree relatives (SER Abstract). *Am J Epidemiol* 1992;**136**:1019

6 Bondy ML, Strom SS, Colopy MW, Brown BW, Strong LC. Accuracy of family history of cancer obtained through interviews with relatives of patients with childhood sarcoma. *J Clin Epidemiol* 1994;**47**:89-96.

7 Gierlowski TC, Ron E, Schneider AB. The accuracy of siblings' family history reports of thyroid and other cancers [letter]. *Am J Public Health* 1995;**85**:408-409.

8 Parent ME, Ghadirian P, Lacroix A, Perret C. Accuracy of reports of familial breast cancer in a case control series. *Epidemiology* 1995;**6**:184-186.

9 Novakovic B, Goldstein AM, Tucker MG. Validation of family history of cancer in deceased family members. *J Natl Cancer Inst* 1996;**88**:1492-1493.

10 Slattery ML, Caan BJ, Duncan D, Berry TD, Coates A, Kerber R. A computerized diet history questionnaire for epidemiologic studies. *J Am dietetic Ass* 1994;**94**:761-766.

11 Airewele G, Adatto P, Cuningham J, et al. Family history of cancer in patients with glioma: A validation study of accuracy. *J Natl Cancer Inst* 1998;**90**:543-544.

12 Love RR, Evans AM, Joston DM. The accuracy of patient reports of a family history of cancer. *J Chron Dis* 1985;4:289-293.

13 Douglas FS, O'Dair L, Robinson M, Evans DGR, Lynch SA. The accuracy of diagnoses as reported in families with cancer: a retrospective study. *J Med Genet* 1999;**36**:309-312.

14 Spraycar M. *Stedman's Medical Dictionary*. Baltimore: Williams & Wilkins, 1995;387

15 Harper PS. *Practical Genetic Counselling*. Oxford: Butterworth-Heinemann, 1998;6

16 Kerber RA, Slattery ML. Comparison of self-reported and database-linked family history of cancer. Data in a case-control study. *Am J Epidemiol* 1997;**146**:244-248.

17 Muller H. Familial cancer in Basel: some aspects. In: Muller H, Weber W, eds. *Familial cancer*. Basel: Karger, 1985;1-5.

18 Aitken JF, Youl P, Green A, Maclennan R, Martin NG. Accuracy of case-reported family history of melanoma in Queensland, Australia. *Melanoma Res* 1996;**6**:313-317.

19 Floderus B, Barlow L, Mack TM. Recall bias in subjective reports of familial cancer. *Epidemiology* 1990;**1**:318-321.

20 Brener D, Altwerger J, Capasso M, et al. Effect of patient education on data quality: implications for assessing hereditary cancer risk on prostate patients. *Am J Hum Genet* 1997;**61**:A187

21 Aitken J, Bain C, Ward M, Siskind V, Maclennan R. How accurate is self-reported family history of colorectal cancer? *Am J Epidemiol* 1995;**141**:683-671.

22 Schmidt S, Becher H, Chang-Claude J. Breast cancer risk assessment: use of complete pedigree information and the effect of misspecified ages at diagnosis of affected relatives. *Hum Genet* 1998;**102**:348-356.

23 Evans DGR, Kerr B, Cade D, Hoare E, Hopwood P. Fictitious breast cancer family history. *Lancet* 1996;**348**:1034

24 Kerr B, Foulkes WD, Cade D, et al. False family history of breast cancer in the family cancer clinic. *Eur J Surg Oncol* 1998;**24**:275-279.

25 Novakovic B, Goldstein AM, Wexler LH, Tucker MA. Increased risk for neuroectodermal tumours and stomach cancer in relatives of patients with Ewing's sarcoma. *J Natl Cancer Inst* 1994;**86**:1702-1706.

Chapter 4
Monitoring referral to clinical cancer genetics services as a means to help targeting educational programs.

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4.1 Abstract

Background: Monitoring regional referrals to a clinical cancer genetics service may help to identify targets for educational programs, aiming at improved care for hereditary cancer patients and families.

Methods: Details of cancer related referrals to our clinical genetics centre, which serves a population of 2 million, were analyzed retrospectively for each of the 1093 consultees seen between 1987 and 1998. Cancer incidence was monitored by the regional cancer registry. In order to compare the referring sources and correct for differences between local populations, we used early-onset cancer (*i.e.* diagnosed <50 yrs) as a surrogate for the unknown local hereditary cancer incidence. We divided the number of referrals for a particular type of cancer by the numbers of patients diagnosed with that type of early-onset cancer in each of the 17 regional hospitals. Referrals from general practitioners and self-referrals, were divided by the number of early-onset cancer in each of the 11 subregions.

Results: High genetic risks were diagnosed in 79 % of the referred families. Breast and ovarian cancer referral ratios differed between regional hospitals (range 0-27 %, $p<0.0001$), between subregional general practitioners (range 2-15 %, $p<0.0001$) and self-referral (range 2-9 %, $p=0.01$). High or low referrals for these tumours from regional specialists were compensated by referrals from others in a minority of subregions. For gastric, colorectal and endometrial cancer, for which the number of referrals were much lower, only subregional differences in self-referral were significant (range 0-17 %, $p=0.03$).

Conclusions: monitoring referral for cancer related clinical genetic services may help in improving the use of these services by identifying potential targets for educational activities.

4.2 Introduction

Hereditary cancer is now thought to account for approximately 1 to 10 % of the total cancer burden[1,2]. DNA tests for hereditary cancer predisposition are increasingly becoming available. Because physicians, patients and advocacy groups, as well as the general public have become aware of these facts, interest in cancer prevention through identification of high-risk individuals by focusing on age of onset of cancer, family history and by using DNA testing has grown[3-8]. This growing interest has caused a rise in the numbers of referrals for genetic diagnosis (including pedigree analysis, physical examination, DNA testing and risk calculation) and counselling. In our country, clinical genetics services are offered equally to all regional hospitals and general practitioners. However, in view of the rapidly evolving indications for genetic analysis, it can be questioned whether doctors will always refer patients appropriately. To improve health care with respect to hereditary cancer, it is important to identify differences in knowledge, so that educational activities can be better directed. These differences may present as dissimilarities in the use of clinical genetics services. Therefore we have retrospectively analyzed the origin of all cancer-related referrals to our clinical genetics centre over the last 11 years.

4.3 Patients & Methods

4.3.1 Organizational background

The Clinical Genetics Centre in Groningen is the provider of clinical genetic services to the north-eastern part of The Netherlands, with a total of 2 million inhabitants. This region is divided into 11 subregions, marked as socioeconomically homogeneous areas by the Netherlands Central Bureau of Statistics. In our centre, clinical genetic analysis and DNA testing is offered for a wide range of disorders including hereditary breast-ovarian cancer and hereditary non-polyposis colorectal cancer. There is a close collaboration with the seven other genetics centres in our country, each of them having its own set of DNA tests, which are made available through all of the clinical genetics centres. Genetic services in the Netherlands have been reviewed in more detail elsewhere[9].

Since legal and financial issues involved in clinical genetics and hereditary cancer may differ strongly between countries and may influence referral, the most important ones are briefly mentioned here. DNA testing for hereditary disorders is restricted by national law to the (non-commercial) clinical genetics centres. The costs of clinical genetic services, as well as periodic screening and prophylactic surgery in individuals with a high genetic cancer risk, are covered by the health care insurance companies. Because of a self-imposed restriction, these companies do not ask for disclosure of DNA test results when buying life insurances or disability insurances, unless the amounts to be covered by these are higher than average. Employers are prohibited by law to ask disclosure of information on genetic diseases by their new employees.

4.3.2 Referring sources

Referrals to our centre come from the general practitioners in our region, from specialists working in the Groningen University Hospital, tightly linked to our centre and from the 17 regional hospitals (A-Q). Some of these regional hospitals share the same subregion: A, G and K; B and Q; F,I ,J and P; M and O. Some consultees came directly to our centre ('self-referrals'). When consultees came to our centre because of a family letter sent by our centre or a similar centre outside of our region, then the referring source was coded as clinical geneticist. The referrals from the Netherlands Foundation for the Detection of Hereditary Tumours were coded separately.

4.3.3 Methods of regional transfer of knowledge on clinical cancer genetics services

In order to reach the physicians in our own region with information on cancer related genetic services, we make use of the Comprehensive Cancer Centre North Netherlands (IKN). In addition to its role as a regional cancer registry, it serves to improve regional oncological health care. Among its activities, the IKN organizes meetings with regional specialists to reach consensus on minimal standards for diagnosis and treatment of cancer, publishes this consensus in a handbook sent to all specialists and also organizes monthly visits of cancer specialists to the regional non-university hospitals to advise on oncological issues, which includes advising on referral for genetic analysis. Clinical geneticists from our centre (RHS, JCO) have recently been appointed by the IKN as consultants on clinical cancer genetics

for each of regional hospitals (primarily to be consulted by phone or through mail). Guidelines on a number of hereditary cancer syndromes have been included in the handbook. We tried to reach the general practitioners, by contributing to their post-graduate training courses.

4.3.4 Registration of source and outcome of referral

For each of the consultees seen in our centre, we registered the date and reason for referral and the specialty of the referring physician in a computer database during the last 11 years (1987-1997). The numbers of (self-)referrals related to cancer or other disorders, for each of these years were counted, stratified for type of referring physician, and plotted on a time scale. We counted the type of final genetic diagnosis in families referred in 1997 for the two groups of cancer which are the subject of the majority of referrals: breast/ovarian cancer, because of suspected hereditary breast-ovarian cancer (HBOC) or gastrointestinal/endometrial cancer, because of suspected hereditary non-polyposis colorectal cancer (HNPCC). The final genetic diagnoses were classified as either:

(a) high-risk families, on the basis of meeting clinical criteria and/or the detection of mutant genes for hereditary cancer; or

(b) medium-risk families, not meeting the criteria for hereditary cancer, but still at an increased risk because of the family history, warranting medical surveillance (albeit sometimes less frequently than in the high-risk families); or

(c) low-risk families, not at a significantly increased genetic risk.

4.3.5 Regional cancer registration

As of 1989, the Comprehensive Cancer Centre North Netherlands (IKN) maintains the cancer registry for the region, identical to the service area of our Clinical Genetics Centre. Details on this cancer registration have been published elsewhere[10]. As part of the registration, data are collected on the number, age, and type of cancer diagnosed in each of the regional hospitals (A-Q).

4.3.6 Correcting number of referrals for population differences

For differences in referral between regional hospitals and between subregions, we restricted ourselves to the most frequent types of referrals, being those for suspected HBOC and HNPCC. The period 1993-1997 was analyzed, because the number of referrals for the relevant cancer types before 1993 were very low. The regional hospitals differ significantly in the type and number of cancer patients seen and possibly also in the number of hereditary cases. Therefore differences in referrals from these hospitals to our centre may simply reflect the differences between the populations they serve. The total number of hereditary cancer cases in these populations is presently unknown. In order to correct for differences between the local populations, we used early-onset cancer (*i.e.* diagnosed <50 yrs) as a surrogate for the unknown local hereditary cancer incidence. We divided the number of breast or ovarian cancer, or gastric, colorectal or endometrial cancer related referrals made in the period 1993-1997 from each of these hospitals, by the number of all patients diagnosed in those hospitals at an age below 50 years with these tumours (as early-onset cancer is a typical characteristic of hereditary cancer) in the same period. This provided us with a referral ratio for each hospital. Referrals from general practitioners and self-referrals, were divided by the number of early-onset cancer in each of the 11 subregions.

If multiple members from one family were referred to our centre by a single source, then only one referral was included in the calculation of the referral ratios.

Some 30% of the cancer related referrals to our centre concern people that do not have cancer themselves but are 'healthy relatives with a strong family history'. For these we could not correct since the number of such persons living in each subregion or being seen in regional hospitals is unknown. However, families in our region show relatively little migration and therefore correcting for hospital and subregional differences in the number of early-onset cancer patients, can be assumed to indirectly correct for the differences in number of healthy individuals with a positive family history of early-onset cancer as well.

4.3.7 University hospital

Sometimes regional physicians referred to our centre through specialists from our university hospital instead of doing so directly. An example of the latter type of referral would be the referral of a woman at high genetic risk to develop breast cancer to a surgical oncologist of the university hospital who in turn referred her for

further genetic analysis. A high or low number of these indirect type of referrals might have compensated a high or low number of direct referrals from a particular subregion. In order to take this possibly compensatory effect into account, we calculated the referral ratios for patients out of the different subregions referred by specialists from our own university hospital.

4.3.8 Statistical analysis

Statistical analysis was performed using the chi-square test from a statistical software package (S.P.S.S. for Windows 6.1.2. , © SPSS, Inc.). Only p-values below 0.05 were considered significant.

4.4 Results

4.4.1 Total number of referrals and type of referring physiscians

The number of cancer related referrals as well as the other referrals between 1987-1997 are shown in figure 1. The proportion of referrals related to cancer increased from 3 % (11 out of 361) in 1987 to 35 % (392 out of 1120) in 1997 (trend $p<0.0001$). There has been a clear rise (trend $p<0.0001$) in the cancer related referrals from general practitioners, surgeons, gynaecologists, internists, and clinical geneticists as well as from the consultees themselves, as shown in figure 2. Referrals from other specialists, including neurologists, dermatologists, ophthalmologists and paediatricians, who mainly referred patients for the rarer tumour syndromes, such as tuberous sclerosis, von Hippel-Lindau disease, retinoblastoma and neurofibromatosis, remained stable.

4.4.2 Outcome of referral in 1997

The type of genetic diagnosis made in the families referred for breast/ovarian cancer and gastrointestinal/endometrial cancer in 1997 was as follows: 79 % high-risk families, 19 % medium-risk families and 2 % low-risk families. These percentages did not differ between the referring sources (specialists, general practitioners and self-referral) ($p=0.17$).

4.4.3 Comparing regional hospitals, subregional general practitioners and self-referral

The ratio of referrals for breast/ovarian cancer and gastrointestinal/endometrial cancer from each of the 17 regional hospitals (A-Q) are shown in figure 3. The number of counted referrals (i.e. max. 1 per family per hospital) were 124 for breast and ovarian cancer and 45 for gastrointestinal/endometrial cancer. The referral ratios showed significant differences for breast and ovarian cancer, ranging from 0-26.8 % ($p<0.0001$). Although the referral ratios for gastrointestinal/endometrial cancer varied markedly from 0 to 30 %, confidence intervals were wider due to the lower number of referrals and referral ratios for these tumours were not significantly different ($p=0.11$).

In order to investigate whether other referral sources might compensate for the variation in the regional hospital referrals, we checked for other referrals in the hospitals' subregion. This appeared to be the case for hospital B,E, H and J (with a low HBOC referral pattern) and for hospital O and P (with a high HBOC referral pattern)(figure 4a). For HNPCC(-like) referrals a similar pattern was seen for hospital C, G,J, K and O (low referral) and F, P and Q (high rėferral) but not for E, H, I and N (figure 4b) .

The total number of counted referrals (*i.e.* max. 1 per family per physician or university hospital) by the general practitioners, the university hospital and for self-referral, were 292 for breast and ovarian cancer and 98 for gastrointestinal and endometrial cancer.

There was a significant inter-subregional difference between the proportion of breast/ovarian cancer related referral for general practitioners (ratios ranging from 2-15 %, $p<0.0001$), self-referral (2-9 %, $p=0.01$) or the university hospital (1-8 %, $p=0.002$) between the different subregions. For gastrointestinal/endometrial cancer, the ratios differed significantly between the subregions for the self-referral category (ratios ranging from 0-17 %, $p=0.03$) but not for general practitioners (0-12 %, $p=0.15$) and the university hospital (0-11 %, $p=0.07$).

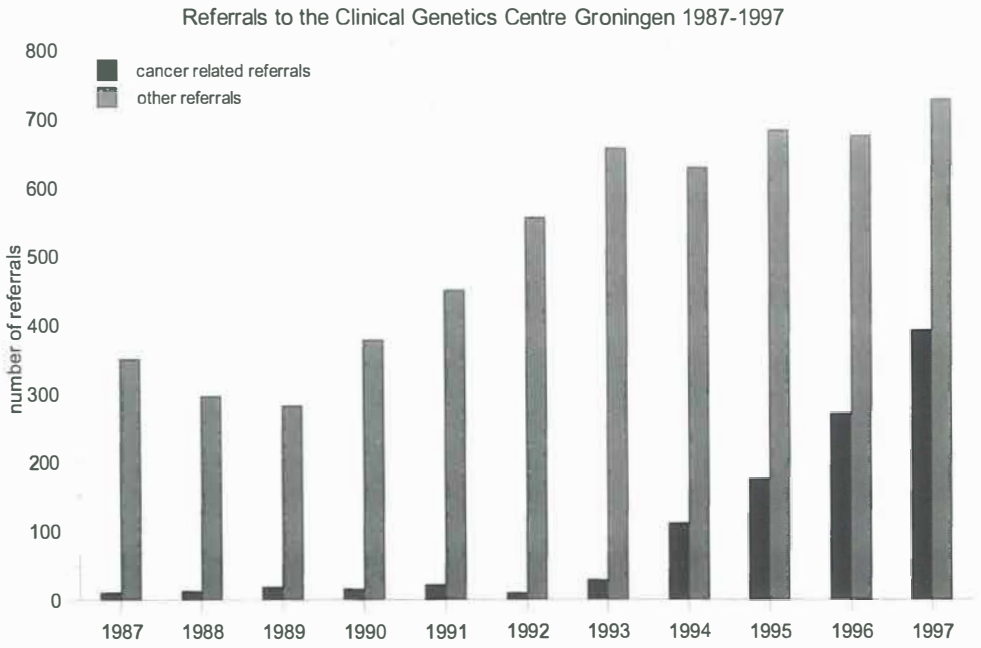

Figure 1 *The number of cancer related referrals as well as the number of referrals for other disorders are shown for each year. After the identification of the genes for hereditary breast-ovarian cancer and hereditary non-polyposis colorectal cancer (HNPCC) in 1993 and 1994, the number of cancer related referrals started to increase rapidly.*

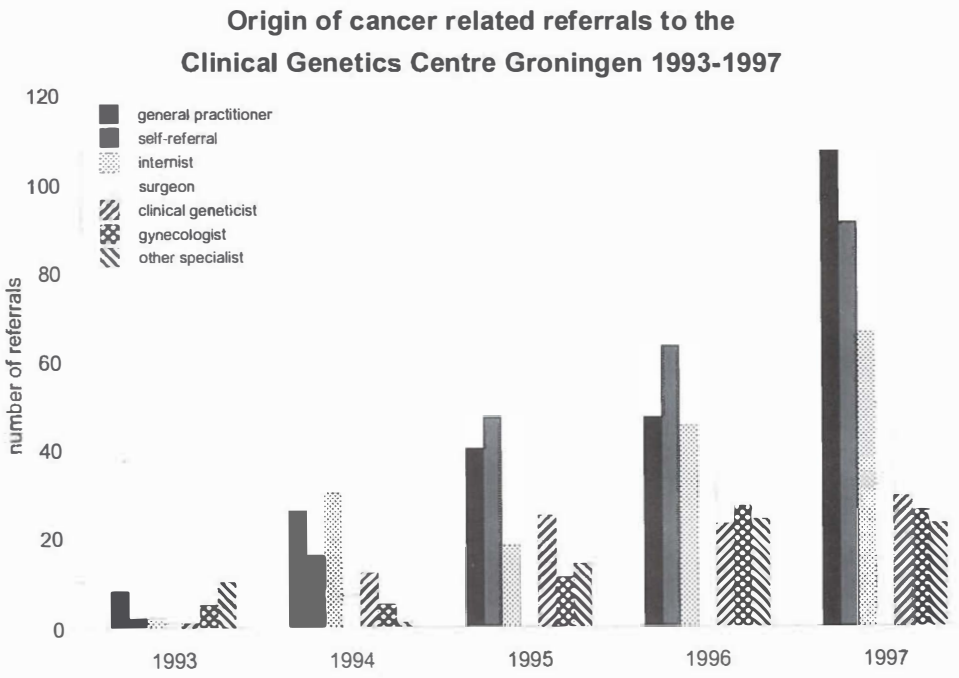

Figure 2 *The number of cancer related referrals are shown for each type of referral (physicians and self-referrals). The number of referrals before1993 are too low for statistical analysis and are not shown.*

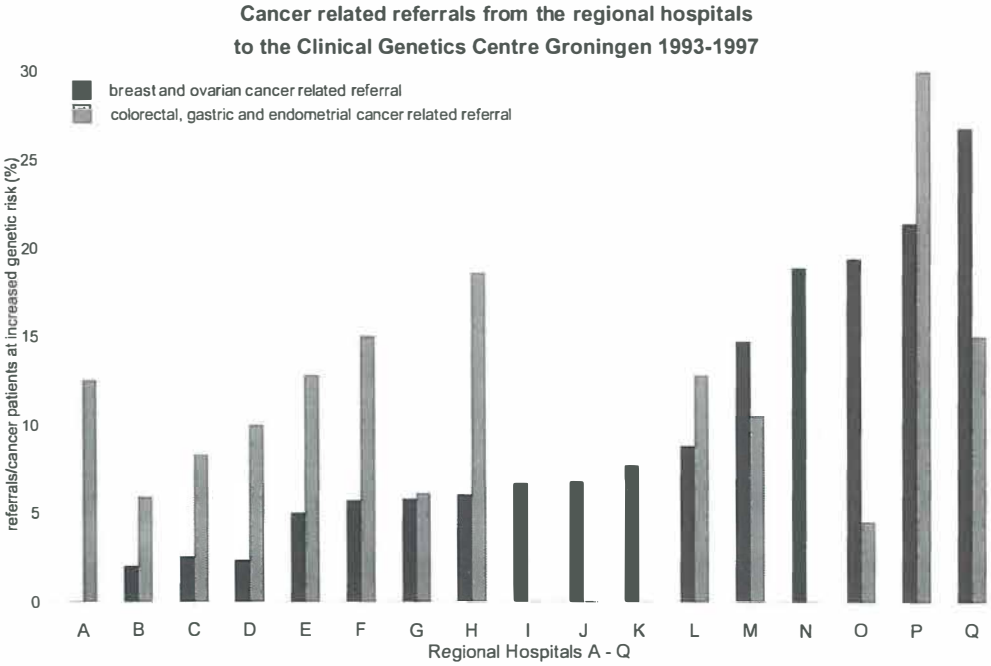

Figure 3 *Referrals from the regional hospitals (A to Q) for breast or ovarian cancer and colorectal, gastric or endometrial cancer related genetic services, respectively. These hospitals differ significantly in the number and type of cancer patients seen, which in itself might explain differences in referral. Therefore, the number of referrals from each hospital were adjusted by dividing it by the number of early-onset (<50 years) cancer patients diagnosed in that hospital between 1993 and 1997. A maximum of one referral per family per hospital was counted. The resulting ratios for breast and ovarian cancer related referrals showed a significant heterogeneity ($p<0.0001$). Although the referral ratios for gastrointestinal and endometrial cancer varied markedly from 0 to 30 %, confidence intervals were wider due to the lower number of counted referrals (45, versus 124 for breast/ovarian cancer) and referral ratios for these tumours were not significantly different ($p=0.11$).*

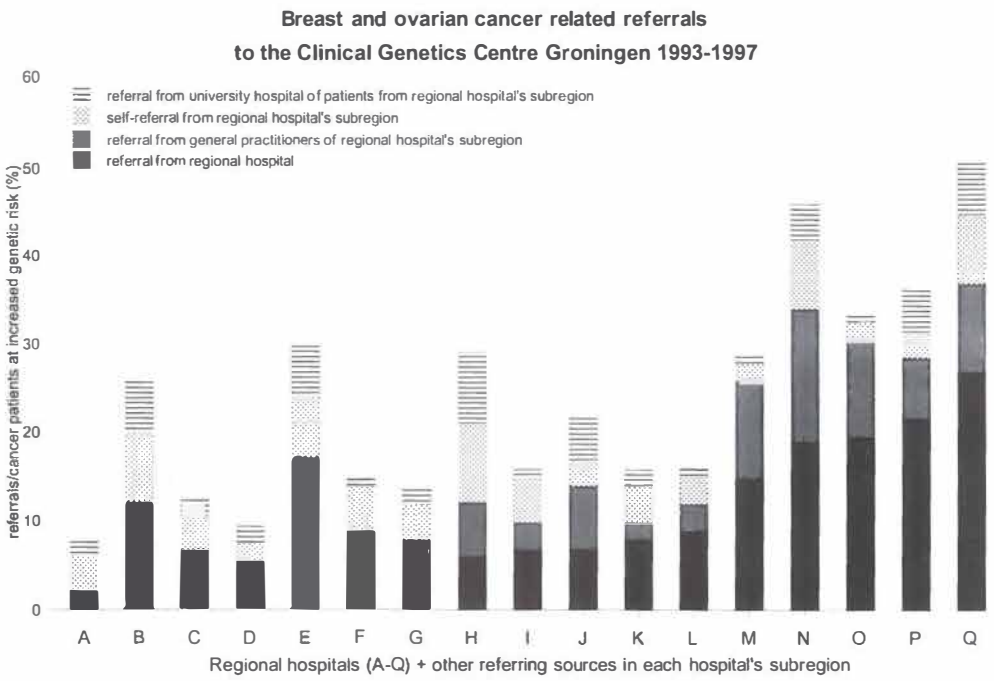

Figure 4a *This figure illustrates the referral for breast or ovarian cancer related genetic services from the regional hospitals (A-Q) and from the general practitioners and consultees themselves (self-referral) from each of the regional hospitals' subregions. Referral ratios of the university hospital of patients from each of the regional hospitals' subregions are also shown. It illustrates that low or high referral rates from the regional hospitals may be compensated by other referring parties in the same subregion.*

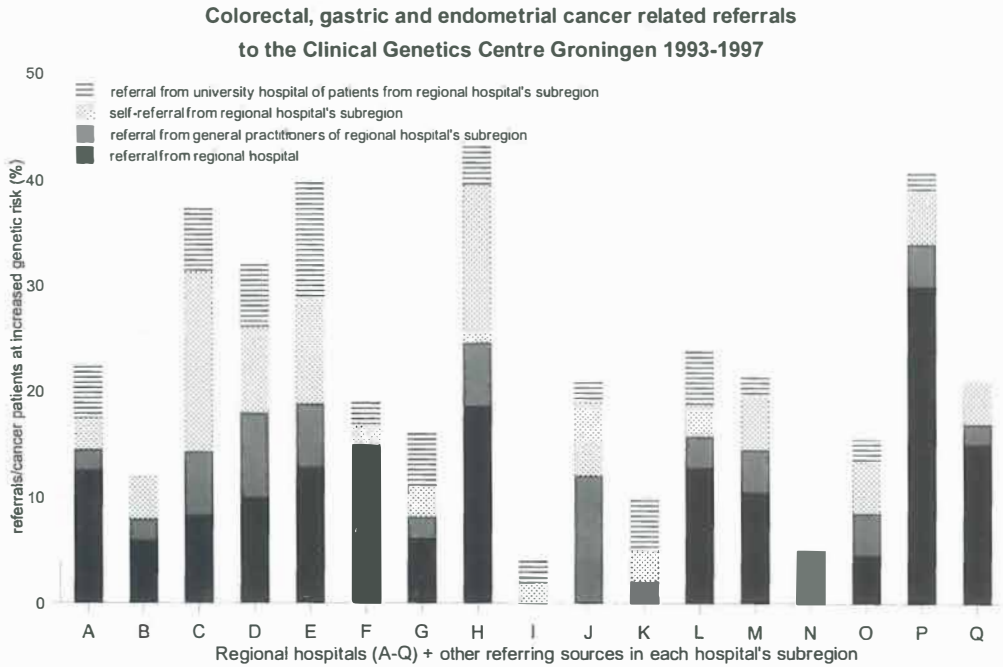

Figure 4b *This figure illustrates the referral for colorectal, gastric or endometrial cancer related genetic services from the regional hospitals (A-Q) and from the general practitioners and consultees themselves (self-referral) from each of the regional hospitals' subregions. Referral ratios of the university hospital of patients from each of the regional hospitals' subregions are also shown.It illustrates that low or high referral rates from the regional hospitals may be compensated by other referring parties in the same subregion.*

4.5 Discussion

The present study shows that our centre has seen a more than 30-fold increase in cancer related genetic counselling over the last eleven years. Similar increases have been observed nation-wide (source: annual reports from the clinical genetics centres). This fast growing demand for genetic analysis and counselling does not necessarily reflect an equal use of these services by the regional physicians and general public. Our study demonstrated that referrals from some of our hospitals and subregions were strikingly lower than from others. As most referrals appear to be warranted, these differences are not explained by differences in unwarranted referrals, but rather hint at an under-recognition of hereditary cancer in those hospitals and areas. The differences suggest that despite all efforts, including the periodic visits of the team of academic specialists to the regional hospitals and contributions to the post-graduate training of general practitioners, information on hereditary cancer so far may not have been adequately provided and 'consumed'.

However, differences in referral should be interpreted with caution, as they are not necessarily a good measure of quality of knowledge on hereditary cancer and genetic services. A referral is the endpoint of the occurrence of (possibly) hereditary cancer in a family, its recognition by the consultee and the physician (or just the consultee) and the decision to consult the clinical genetics centre because of this. There may be several valid reasons for not seeking genetic analysis in individual cases. In addition, some physicians, especially those working near the south and western borders of our region may have referred to other clinical genetics centres or (for pedigree analysis) to the Netherlands Foundation for the Detection of Hereditary Tumours. Calculating referral ratios correcting the number of referrals for the population diagnosed with early-onset cancer is only an indirect correction for possible regional differences in hereditary cancer incidence. Also, the number of healthy relatives with a family history of cancer seen by the regional specialists and general practitioners is presently unknown.

Our monitoring study could not directly reveal under-recognition of hereditary cancer. Rather, it aimed at collecting data on referral to start and support a dialogue on clinical cancer genetics services with the different hospitals and other groups of physicians, and offer 'customized' educational updates if necessary.

We will continue monitoring referral and it will be interesting to observe if any changes in referral patterns will unfold. The need to make such an audit of regional access to highly specialized cancer consultation services as part of a quality-assurance program has recently be stressed[11]. In addition to the education of physicians on new developments in genetics, education of cancer patients and the general public on these issues is important to clarify the potential of genetic testing as well as its limitations. Educational meetings have been and will be organized in many of our subregions for this purpose.

We conclude that the approach we described to assess and monitor regional referrals to clinical cancer genetics services is potentially useful, because it may reveal unequal distribution of genetic health care and facilitate the targeting of educational programs. We assume that further refinement of our model may be necessary to be useful on a larger scale. The ultimate goal of these efforts is to ensure that cancer patients and their families benefit maximally from the new developments in clinical cancer genetics.

4.6 Acknowledgements

We thank Marijke Mathot for database management and Jennita Reefhuis and Hermien de Walle for help in the statistical analysis.

4.7 References

1 Ponder BAJ. Inherited predisposition to cancer. *Trends Genet* 1990;**7**:213-218.

2 Fearon ER. Human cancer syndromes: Clues to the origin and nature of cancer. *Science* 1997;**278**:1043-1050.

3 Croyle RT, Lerman C. Interest in genetic testing for colon cancer susceptibility: cognitive and emotional correlates. *Prev Med* 1993;**22**:284-292.

4 Struewing JP, Lerman C, Kase RG, Giambarresi TR, Tucker MA. Anticipated uptake and impact of genetic testing in hereditary breast and ovarian cancer families. *Cancer Epidemiol Biomarkers Prev* 1995;**4**:169-173.

5 Evans DGR, Maher ER, Macleod R, Davies DR, Craufurd D. Uptake of genetic testing for cancer predisposition. *J Med Genet* 1997;**34**:746-748.

6 Andrykowski MA, Lightner R, Studts JL, Munn RK. Hereditary cancer risk notification and testing: How interested is the general population? *J Clin Oncol* 1997;**15**:2139-2148.

7 Julian-Reynier C, Eisinger F, Vennin P, et al. Attitudes towards cancer predictive testing and transmission of information to the family. *J Med Genet* 1996;**33**:731-736.

8 Tambor ES, Rimer BK, Strigo TS. Genetic testing for breast cancer susceptibility: Awareness and interest among women in the general population. *Am J Med Genet* 1997;**68**:43-49.

9 ten Kate LP. Genetic services in the Netherlands. *Eur J Hum Genet* 1997;**5**:125-129.

10 Schaapveld M, Otter R. *Results of cancer registration in the Northern part of the Netherlands 1989-1994.* Groningen: Dijkhuizen van Zanten, 1998;6-10

11 Kunkler I. European breast-cancer meeting [Letter]. *Lancet* 1998;**352**:1711

Chapter 5
Familial Cancer Database: a clinical *aide-mémoire.*

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5.1 Abstract

Cancer is associated with a wide range of hereditary disorders. Recognizing these disorders in cancer patients may be of great importance for the medical management of both patients and their relatives. Conversely, recognizing the fact that cancer may develop in patients already diagnosed with a hereditary disorder may be important for the same reason. We have developed a stand-alone interactive computer program, Familial Cancer Database, to assist the clinician in making a genetic differential diagnosis in cancer patients as well as in becoming aware of the tumour spectrum associated with a particular hereditary disorder already diagnosed. The program tries to match tumour and non-tumour features observed in patients and their families with any of the more than 300 disorders presently contained in the database and provides a clinical synopsis for each of these disorders. Familial Cancer Database is offered as part of the Familial Cancer and Prevention project of the UICC Cancer Epidemiology and Prevention program. The software can be downloaded from the internet at http:/www.uicc.ch/ [download-site will be activated at the time of the journal publication of this paper]

The contents of this paper and the Familial Cancer Database are solely the responsibility of the authors and do not necessarily reflect the official views of the UICC.

5.2 Introduction

As a result of recent clinical and basic research on hereditary disorders, there has been a vast increase in knowledge on hereditary aspects of cancer[1]. Cancer has been reported as a feature of more than 200 hereditary disorders[2]. These disorders include the human cancer syndromes, associated with high life-time risks to develop cancer, as well as with a much larger group of disorders associated with a smaller increase in cancer risk. The recognition of a hereditary nature of cancer in patients by their physicians may have major implications for both treatment and cancer prevention, not only for those patients, but also for their families[3]. From a different clinical perspective, recognizing the fact that tumours may be part of the clinical spectrum of hereditary disorders already diagnosed, may lead to early detection or prevention of those tumours.

Some hereditary tumour syndromes are well known and easily recognized by the majority of physicians, *e.g.* the classical phenotype of familial adenomatous polyposis. Others are less well known. The same is true for oncological aspects of hereditary disorders already diagnosed in non-cancer patients. In some, the fact that cancer may complicate the disease is well known, *e.g.* in Recklinghausen disease or Beckwith-Wiedemann syndrome. In others, it is not. Moreover, it may be difficult for physicians to keep an overview of cancer-associated hereditary disorders, because new data on these disorders are continuously being published, which leads to new clinical classifications and diagnostic criteria. With this in mind, we set out to develop an updatable computer program to assist the physician in recognizing hereditary disorders in cancer patients as well as in recognizing the possibility of cancer developing in their patients already diagnosed with a hereditary disorder.

5.3 Material and Methods

A list of proven or presumed hereditary disorders associated with tumour development was compiled from the literature by consulting the following sources: McKusick's on-line catalogue of hereditary phenotypes (Online Mendelian Inheritance in Man, OMIM, found at www.ncbi.nlm.nih.gov/Omim), a catalogue of human genes predisposing to neoplasia [4], textbooks on hereditary cancer[5-14], as of 1994, weekly searches in Current Contents on Diskette, additional searches in Medline, and literature references found through the previously mentioned sources. The same was done for congenital anomalies (not necessarily hereditary) and

constitutional chromosomal abnormalities (*e.g.* Down syndrome) associated with cancer, because these disorders are considered in a clinical genetic differential diagnosis as well.

All disorders thus identified were stored in a relational database, written in Microsoft Access 1.1(© Microsoft Corporation, Inc.). Although the emphasis of this database is on malignant tumours, hereditary disorders which feature (multiple) benign tumours with a potential risk for malignant transformation were included as well.

For each of the disorders included in the database, information was stored on 9 items: name, synonyms, mode of inheritance, associated genes, McKusick's number, tumour spectrum, non-tumour features, a clinical summary and references. Inclusion criteria for these items were as follows. The McKusick number, mode of inheritance and genes involved were taken from OMIM, updated from more recent literature if available. In cases of doubt with regard to modes of inheritance, as expressed by the catalogue and/or the literature, a question mark was added to these entries in the database.

Tumours associated with the included disorders were labelled with regard to the strength of those associations. Tumours which, based on epidemiological data or molecular studies, were significantly associated with a particular disorder were labelled 'certain'. In many of the rare disorders where no such statistical or molecular data were available, we included tumour types shared by all or the majority of patients reported with that particular disorder. As a general rule, we also included tumours with a relatively early age at diagnosis. In case of doubt, tumours were labelled as 'possible' association. Of the non-tumour features, only those that are most characteristic for each disorder, according to textbooks and review articles, were included. The clinical summary contains a general description of the disorder, mentioning available diagnostic criteria and risk figures. References were included to support the clinical summaries, in particular with regard to oncological and genetic aspects.

The graphical user-interface to access the contents of the database was written in Microsoft Visual Basic Pro 3.0 (© Microsoft Corporation, Inc.) in conjunction with TrueGrid 2.1 Pro (© Apex Software Corporation). By deliberately using 16-bit versions of Access, TrueGrid and Visual Basic, minimal system requirements to run Familial Cancer Database were kept modest: IBM compatible personal computer with a 80486 processor (or better), at least 4 megabytes of internal memory

(RAM), a mouse, SVGA screen and the Windows 3.1 (or higher) operating system (© Microsoft Corporation, Inc.).

The program was tested in the clinic by a panel representing the various medical disciplines involved in the diagnosis and treatment of cancer patients and of patients with hereditary disorders (see: acknowledgements). The panel's feed-back was used to refine the database contents and the user-interface.

5.4 Results

At the time of submission of this paper, the database contained information on 307 disorders, approximately 90 % of which were proven or presumed hereditary disorders, the others being congenital anomalies and constitutional chromosomal aberrations. The interface was programmed to present four main options to the user: 'Find Syndromes', 'Browse Syndromes', 'Browse References' and 'Browse Chromosomes/Genes'. The first two options are the key features of the program.

The 'Find Syndromes' option allows the physician to draw up a search profile choosing from 425 different benign and malignant tumour types and 450 non-tumour features (*e.g.* immune deficiency) using an organ-oriented interface (figure 1). Because we wanted the program to be able to deal with family histories when those had not yet been verified and therefore possibly would be less detailed in describing the specific tumour types, several broad search terms were included. Examples of these are gastrointestinal cancer, brain tumour and uterine tumour. On running the search profile, the program tries to match it with the disease profiles stored in the database and lists the result (figure 2). The details of the listed disorders can subsequently be viewed (figure 3). For each of the tumour types listed in the search profile, the user may choose to have the program search for disorders certainly or possibly featuring that tumour in its tumour spectrum ('wide' search mode) or only certainly doing so ('narrow' search mode).

In the 'Browse Syndromes' option, the disorder of interest to the user can be selected from an alphabetical listing of all disorders included in the database and can subsequently be viewed for details (figure 3).

In the 'Browse Chromosomes/Genes' option, the user can select a specific chromosomal area and view the genes located in that area, which are associated with cancer risk if mutated in the germline. Also, the disorders associated with any of the 171 genes included in the database can be viewed.

Finally, the 'Browse References' option allows the user to search the more than 1850 references in the database by name of the first six authors. An on-line help option is available to the user, explaining all the features of the program, its limitations and providing general information on hereditary disorders and hereditary cancer.

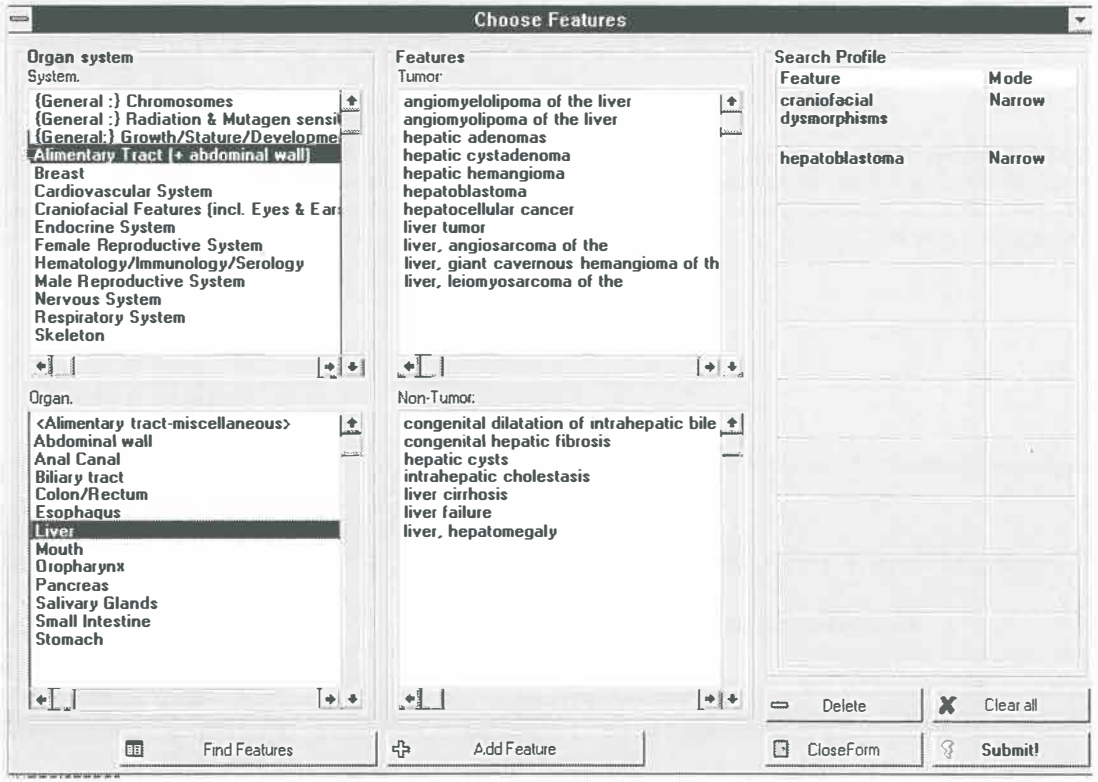

Figure 1 *By selecting tumour types and (if relevant) non-tumour features from the organ-oriented listings in this screen, the user can compose a search profile reflecting the features observed in a patient and his or her relatives. Running the profile will make the program try to match the features from the profile with those of the disorders stored in the database.*

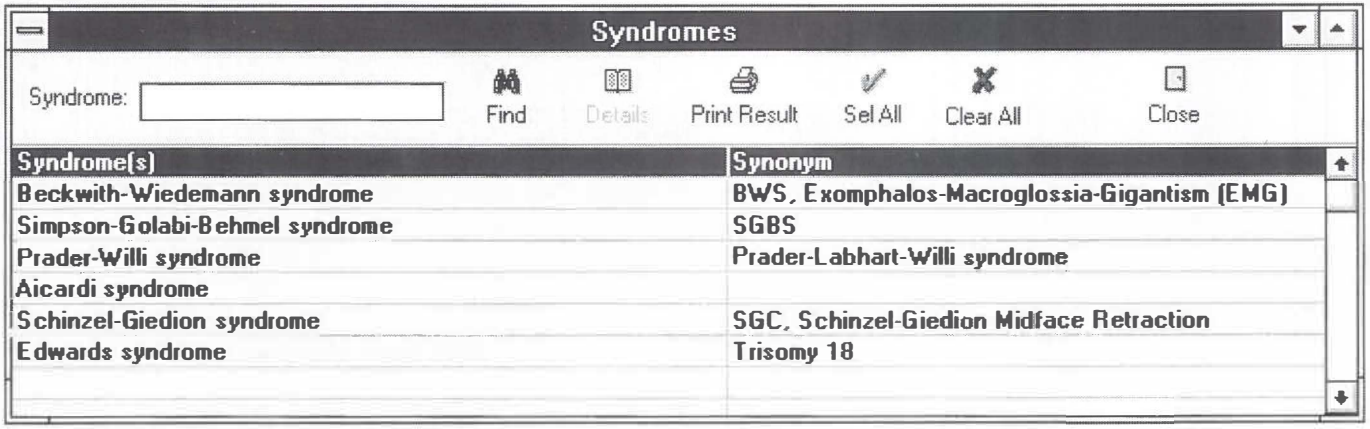

Figure 2 *This screen displays the disorders which manifest either certainly or possibly (i.e. using the 'wide' search mode) hepatoblastoma as well as craniofacial dysmorphisms. If the search profile had been run in the search mode 'narrow', i.e. matching only with disorders of which we are certain that they include both features in their phenotype, then only Beckwith-Wiedemann syndrome would have come up. The clinical details as presented by the program for one of the listed disorders, Aicardi syndrome, is shown in figure 3.*

Name: Aicardi syndrome

---> Mode of Inheritance: XLD

---> Genes: AIC#, located on Xp22.3

---> McKusick number(s): 304050

---> Tumor Spectrum:
angiosarcoma (possible feature)
choroid plexus papilloma (possible feature)
colorectal polyps (possible feature)
gastric polyps (possible feature)
hepatoblastoma (possible feature)
lipoma (possible feature)
palate, benign teratoma of (possible feature)
parapharyngeal embryonal cell cancer (possible feature)

---> Non-Tumor Features:
chorioretinal lacunae
cleft lip
cleft palate
corpus callosum agenesis
facies, asymmetric
gross motor delay
mental deficiency
plagiocephaly
rib anomalies
seizures
vertebral anomalies

--->Comment:
Clinical hallmarks are infantile spasms, corpus callosal agenesis, chorioretinal abnormalities (lacunae) and severe psychomotor retardation. Other findings include plagiocephaly, facial asymmetry, cleft lip and palate and costovertebral anomalies. A number of tumors have been reported in association with Aicardi syndrome[1-5]: choroid plexus papilloma (the most frequent tumor), gastric hyperplastic polyps, rectal polyp, soft palate benign teratoma, hepatoblastoma, parapharyngeal embryonal cell cancer; limb angiosarcoma and scalp lipoma.

--->References:
1) Robinow M, Johnson GF, Minella PA. Aicardi syndrome, papilloma of the choroid plexus, cleft lip, and cleft of the posterior palate. J Pediat 1984;104(3):404-5.
2) Tagawa T, Mimaki T, Ono J, Tanaka J, Imai K, Yabuuchi H. Aicardi syndrome associated with an embryonal carcinoma. Pediat Neurol 1989;5(1):45-7.
3) Tanaka T, Takakura H, Takashima S, Kodama T, Hasegawa H. A rare case of Aicardi syndrome with severe brain malformation and hepatoblastoma. Brain Dev 1985;7:507-12.
4) Trifiletti RR, Incorpora G, Polizzi A, Cocuzza MD, Bolan EA, Parano E. Aicardi syndrome with multiple tumors: a case report with literature review. Brain Dev 1995;17:283-5.
5) Tsao CY, Sommer A, Hamoudi AB. Aicardi syndrome, metastatic angiosarcoma of the leg, and scalp lipoma. Am J Med Genet 1993;45:594-6.

Figure 3

Example of a clinical details screen. The '#' symbol after the gene's name means this gene has not been cloned yet.

5.5 Discussion

We have developed a computer program to help clinicians to orientate themselves on the possible hereditary aspects of tumours in their patients, as well as on oncological aspects of hereditary disorders already diagnosed in their patients. Because knowledge of these aspects rapidly changes, the use of software offers an advantage over printed literature as it can be more readily updated and redistributed (*e.g.* through the Internet). For orientation purposes it also offers advantages over on-line computerized literature searches because it offers information in a review format. Online Mendelian Inheritance in Man (OMIM) is a valuable and updated source of information on hereditary disorders in a review format. However, it is not specifically focused on oncological aspects of these disorders.

Our program has of course limitations as well. The user needs a computer and (basic) knowledge of computers to use the software. Therefore, depending on the circumstances, consulting textbooks or other printed sources may be more convenient. Also, the present version of our program does not contain any illustrations and because of its focus on the genetic differential diagnosis of tumours, it does not discuss clinical management of disorders and covers only the most characteristic non-oncological aspects.

A limitation for any type of review on hereditary disorders, congenital anomalies and constitutional chromosomal abnormalities stems from the fact that for many of these disorders and for a wide range of tumours, there is no solid statistical or molecular evidence yet that these tumours are truly part of the phenotype. We have explicitly labelled uncertain associations as 'possible' in the database and the user may choose to have the program search only for certain associations. Still, we are aware of the fact that deciding on the strength of association of a tumour type with a particular disorder in the absence of statistical or molecular proof is often arbitrary.

The clinical value of searching databases as well as printed literature does not depend on the data contained in those sources alone. The skill of the user in selecting good 'search handles' from the various features observed in a patient and family is important as well. Selecting tumours or features less suggestive of a hereditary cancer predisposition, *e.g.* a case of lung cancer diagnosed in a 70-year-old heavy smoker from a family with early-onset ovarian cancer, may very well devaluate the search results. Although the on-line help-function of the program gives general hints on selection and discusses modes of inheritance, the user

should have a certain knowledge of genetics and oncology to make good use of the program.

Our program is primarily aimed at physicians and genetic counsellors working in familial cancer clinics and in departments of medical genetics, oncology (surgical and medical disciplines), pathology and paediatrics. To determine the practical value of the program and allow it to further mature, it needs to be tested by a large group of users from various medical disciplines over some longer period of time. We intend to perform an user survey once this group is sufficiently large. The Familial Cancer Database is offered free of charge as part of the Familial Cancer and Prevention project of the UICC Cancer Epidemiology and Prevention program. The software can be downloaded from the Internet at http:/www.uicc.ch/[site will be activated at the time of the journal publication]. The authors welcome comments on all aspects of the program at facd@med.rug.nl and will continue to update, refine and expand it as warranted from users' feed-back and scientific, clinical and technological developments.

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Asperen CJ v , Burgt I vd, Castedo S, Jonkman M, Kleibeuker JH, Kluijt I, Maher ER, Menko F, Meijers H, Molenaar WM, Moog U, Mourits M, Mueller Hj, Oosterwijk JC, Otter R, Passini B, Szanto J, Utsonomiya J, Vasen HFA, Verhoef S, Vries EGE de, Weber W.

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5.7 References

1 Fearon ER. Human cancer syndromes: Clues to the origin and nature of cancer. *Science* 1997;**278**:1043-1050.

2 Lynch HT, Fusaro RM, Lynch J. Hereditary cancer in adults. *Cancer Detect Prev* 1995;**19**:219-233.

3 Parry DM, Mulvihill JJ, Miller RW, Berg K, Carter CL. Strategies for controlling cancer through genetics. *Cancer Res* 1987;**47**:6814-6817.

4 Mulvihill JJ, Davis S, Fromkin KR. The catalog of human genes predisposing to neoplasia. In: Weber W, Mulvihill JJ, Narod SA, eds. *Familial cancer management*. New York: CRC press, 1996;203-237.

5 Lynch HT. *Cancer genetics*. Springfield: Charles C Thomas Publisher, 1976

6 Mulvihill JJ, Miller RW, Fraumeni jr JF. *Genetics of human cancer*. New York: Raven Press, 1977

7 Muller H, Weber W. *Familial cancer*. Basel: Karger, 1985;

8 Chaganti RSK, German J. *Genetics in Clinical Oncology*. New York/Oxford: Oxford umiversity press, 1985;

9 Hodgson SV, Maher ER. *A practical guide to human cancer genetics*. Cambridge: Cambridge University Press, 1993;

10 Ponder BAJ ed. *Genetics of malignant disease*. Edinburgh: Churchill Livingstone, 1994

11 Ponz de Leon M. *Familial and hereditary tumors*. Berlin: Springer-Verlag, 1994

12 Eeles RA, Ponder BAJ, Easton DF, Horwich Ae. *Genetic predisposition to cancer*. London: Chapman & Hall Medical, 1996

13 Vogelstein B, Kinzler KW eds. *The genetic basis of human cancer*. New York: McGraw-Hill, 1998

14 Offit K. *Clinical cancer genetics. Risk counseling & management*.. New York: Wiley-Liss, 1998

Appendix I
RET mutation screening in familial cutaneous lichen amyloidosis and in skin amyloidosis associated with multiple endocrine neoplasia

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In several families, multiple endocrine neoplasia type 2A (MEN 2A) has been found in association with cutaneous lichen amyloidosis. It has been debated, however, whether the skin amyloidosis found in MEN 2A families, localized exclusively in the interscapular area, represents the same anomaly as that found in autosomal dominant familial cutaneous lichen amyloidosis, which is more generalized. We screened two MEN 2A families with associated skin amyloidosis for germline mutations in the *RET* gene responsible for the MEN 2A cancer syndrome, and found the same mutation characteristic of MEN 2A in both families. We also screened probands from three pedigrees with familial cutaneous lichen amyloidosis for *RET* mutations. In none of the *RET* coding and flanking intronic sequences was a mutation detected. This most probably indicates that skin amyloidosis found in some MEN 2A families and familial cutaneous lichen amyloidosis are different conditions. Consequently, patients with apparent familial cutaneous lichen amyloidosis do not appear to be at risk for MEN 2A.
Key words: genetic heterogeneity/clinical heterogeneity/etiologic heterogeneity. J Invest Dermatol 107:215–218, 1996

Cutaneous lichen amyloidosis (CLA) is a rare disorder characterized by deposits of amyloid in the papillary dermis. Sporadic as well as autosomal dominant hereditary forms have been documented. Gagel *et al* (1989) reviewed 63 of these hereditary cases. Here we refer to the hereditary form as familial cutaneous lichen amyloidosis (familial CLA).

CLA-like skin lesions have also been found in patients with multiple endocrine neoplasia type 2A (MEN 2A) (Gagel *et al*, 1989; Nunziata *et al*, 1989; Ferrer *et al*, 1991; Kousseff *et al*, 1991; Chabre *et al*, 1992; Robinson *et al*, 1992; Pacini *et al*, 1993). MEN 2A is a neoplastic syndrome characterized by C-cell hyperplasia, medullary thyroid carcinoma, pheochromocytoma, and parathyroid hyperplasia. The disorder is caused by specific germline mutations in the *RET* proto-oncogene (Donis-Keller *et al*, 1993; Mulligan *et al*, 1993).

Because patients have been found with CLA lesions in several MEN 2A families, it has been suggested that patients having sporadic or familial CLA should be considered at risk for the MEN 2A syndrome and therefore should be tested for MEN 2A mutations (Nunziata *et al*, 1989; Ferrer *et al*, 1991; Chabre *et al*, 1992). Based on the association of both of these conditions, *RET* gene mutations have been thought responsible for the skin amyloidosis found in MEN 2A patients. We therefore screened two MEN 2A families with associated CLA and three families with familial CLA for *RET* germline mutations.

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Abbreviations: CLA, cutaneous lichen amyloidosis; FCLA, familial case of CLA family; MEN 2A/2B, multiple endocrine neoplasia type 2A/type 2B; MEN 2A/CLA, multiple endocrine neoplasia type 2A associated with CLA; SSCP, single-strand conformation polymorphism.

MATERIALS AND METHODS

Familial CLA The three families participating in this study had CLA in at least two generations (**Fig 1**). All affected family members were examined by a dermatologist. Light microscopic and electron microscopic evidence of amyloid was found in skin biopsy specimens from at least two individuals in each of the familial CLA families, FCLA-2 and FCLA-3. Although no electron microscopic analysis was performed on patient material from family FCLA-1, the diagnosis in this family was based on a characteristic clinical picture and on histopathologic and immunofluorescence examination of skin specimens. In all three families, the CLA lesions were found mainly on the arms and legs.

MEN 2A/CLA Families Families MEN 2A/CLA-1 and -2 have been described before. **Figure 2** shows the relevant parts of the pedigrees. Family MEN 2A/CLA-1 has been reported by Kousseff *et al* (1991), who gave a detailed description of the CLA lesions. Family MEN 2A/CLA-2 has been described as family B by Lips *et al* (1994). Some of the patients in this family appeared with lesions in the interscapular region only and were clinically diagnosed as having CLA upon examination by a dermatologist. Light microscopic evaluation of biopsy specimens of the lesions failed, however, to detect amyloid.

Single-Strand Conformation Polymorphism (SSCP) Analysis High-molecular-weight DNA from patients from families MEN 2A/CLA-1, FCLA-1, FCLA-2, and FCLA-3 was used for SSCP analysis of all 20 *RET* exons. DNA amplification was carried out on 150 ng of DNA in 1× Super *Taq* reaction buffer using 0.125 U of Super *Taq* (HT Biotechnology Ltd., Cambridge, UK) in a total volume of 30 μl containing 20 μM

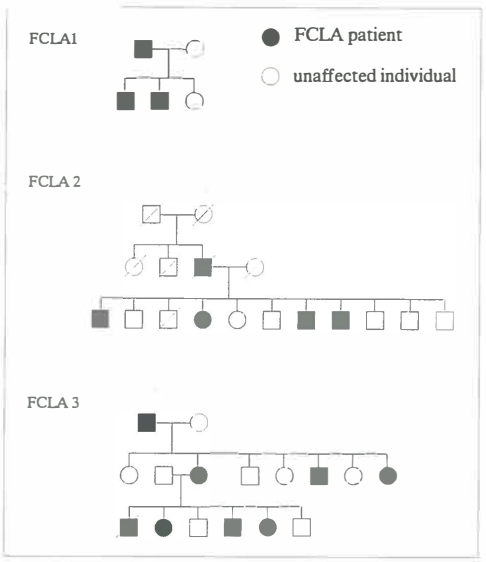

Figure 1. Pedigrees of families with familial cutaneous lichen amyloidosis (FCLA) included in this study. *Solid symbols* indicate individuals affected with CLA.

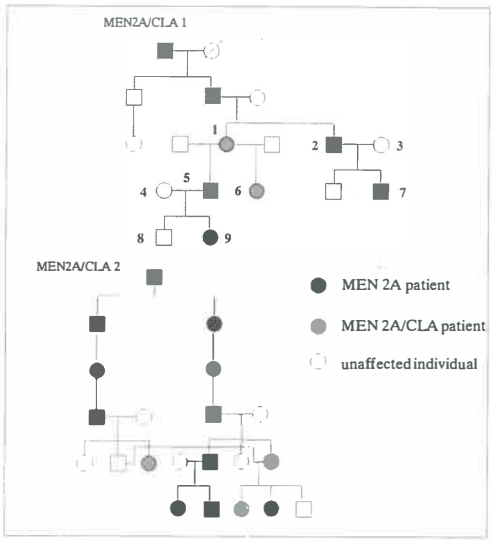

Figure 2. Pedigrees of families with multiple endocrine neoplasia type 2A (MEN 2A) and cutaneous lichen amyloidosis (CLA) included in this study. *Solid symbols* indicate individuals affected with MEN 2A; *shaded symbols* show individuals affected with both MEN 2A and CLA. The numbers in pedigree 1 correspond to those of **Fig 4**.

deoxycitidine triphosphate (dCTP), 200 μM deoxyadenosine triphosphate/thymidine triphosphate/guanosine triphosphate, and 1 μCi of [α-^{32}P]dCTP. The polymerase chain reaction (PCR) consisted of 30 cycles of 92°C for 40 s, 72°C for 60 s, and another 60 s for annealing at the appropriate temperature. **Table I** lists the primers (100 ng of each primer) used for each exon of the *RET* gene, the annealing temperature, and the restriction enzymes used when relatively long PCR products were obtained. Electrophoresis was carried out in a 6% polyacrylamide gel under at least two different conditions. Glycerol concentrations used were 0%, 5%, or 10%, at 4°C, 20°C, or 30°C, respectively. We also used Mutation Detection Enhancer gel solution (At-Biochem, Malvern, PA) as a replacement for acrylamide and glycerol and ran the gels at 30°C in 0.5× Tris-borate buffer at a maximum of 1750 V and 60 W in a temperature-regulated LKB 2010 Macrophore electrophoresis unit (Uppsala, Sweden).

Polymorphisms in the *RET* Gene Intragenic *RET* polymorphisms (see **Table II**) were analyzed by carrying out digestions with the restriction enzymes listed or, in the case of exon 18, by SSCP analysis.

Sequence Analysis For all families, sequence analysis was carried out on exons 10 and 11, which are known to account for more than 95% of the mutations found in MEN 2A (Mulligan *et al*, 1994; Schuffenecker *et al*, 1994). In addition, all SSCP variants observed were sequenced. For SSCP and sequence analysis, the same primer sets were used. For sequence analysis, however, one of the primers of each set was biotinylated. DNA amplification was carried out as described earlier. PCR products were separated in a 2% low melting point agarose gel. After ethidium bromide staining, bands were cut out and isolated with the Sephaglas BandPrep kit (Pharmacia Biotech, Uppsala, Sweden). The two single strands were separated using Dynal beads (DYNAL AS, Oslo, Norway). They were sequenced with the T7 sequencing kit (Pharmacia Biotech) and [α-^{32}P]dCTP. A 6% sequencing gel was used for electrophoresis.

RESULTS

***RET* Mutation Screening in MEN 2A/CLA Families** A search for mutations throughout the entire *RET* gene by means of SSCP revealed in one family (MEN 2A/CLA-1) a conformation variant in exon 11 in all affected family members (MEN 2A and MEN 2A/CLA patients). Upon sequence analysis, this appeared to be caused by transition T1900→C, resulting in the substitution of an arginine for a cysteine at codon 634 (Cys634→Arg) **(Fig 3)**.

Sequence analysis of exons 10 and 11 of the *RET* gene showed the same mutation, T1900→C, in family MEN 2A/CLA-2 in all affected family members (MEN 2A and MEN 2A/CLA patients). Confirmation of the mutation was possible, as this mutation creates a *Hha*I site. Exon-11 alleles with the mutation should have an extra *Hha*I site, giving rise to a PCR product that is 60 bp shorter after digestion. **Figure 4** shows restriction analysis of exon 11 for nine individuals from family MEN 2A/CLA-1. All affected persons indeed showed two bands, the wild-type and the 60-bp–shorter mutant band, whereas nonaffected persons did not have the restriction site and showed only the wild-type band.

***RET* Mutation Screening in Familial CLA Families** SSCP analysis of all exons of the *RET* gene did not show a causative *RET* mutation, nor did sequence analysis of exons 10 and 11.

Haplotyping Using Intragenic Polymorphisms All families were haplotyped for all of the intragenic markers listed in **Table II**. No specific polymorphism seemed to co-segregate with the familial CLA or MEN 2A/CLA phenotypes (data not shown).

DISCUSSION

Phenotypic diversity due to mutations affecting different domains of a gene product is a frequent phenomenon known as allelic heterogeneity. The *RET* gene is a well-known example. Base-pair substitutions affecting one of five highly conserved cysteine residues in the extracellular part of the protein are associated with MEN 2A and familial medullary thyroid carcinoma (Mulligan *et al*, 1994; Schuffenecker *et al*, 1994). Furthermore, a missense mutation substituting threonine for methionine at codon 918 in the tyrosine kinase domain of the protein has been found to be uniquely associated with MEN 2B (Carlson *et al*, 1994; Eng *et al*, 1994; Hofstra *et al*, 1994). Mutations throughout the gene of one of the two alleles, presumably leading to inactivation of the protein, have been found responsible for a proportion of patients suffering from Hirschsprung disease (Edery *et al*, 1994; Romeo *et al*, 1994). The combined occurrence of both MEN 2A and CLA in some families

Table I. Primers Used in Single-Strand Conformation Polymorphism and Sequence Analysis of the *RET* Proto-Oncogene[a]

Exon Number	Forward Primer	Reverse Primer	Annealing Temperature (°C)	Length (bp)	Restriction Enzyme
1	5′-GAGTGCCCCGGAACGTGCGT-3′	5′CGCGTGCCCACGGCAAACAG-3′	68	166	
2	5′-CCATATTCTCACCATCCCTC-3′	5′-AGTGTCAGCGGCTGTGATAA-3′	58	387	*Sau*3A
3	5′-GGACCAGGGTTTACACCAGC-3′	5′-GCTTGTGTCAAGGGCTCGCA-3′	58	466	*Sma*I
4	5′-CCTTCCCGAGGAAACGGCT-3′	5′-CGAACTGTGGCCGGAGACAG-3′	58	423	*Bste*II
5	5′-CCTAAGGTCTCTGGTTTTGG-3′	5′-AAGAGCGACCACCTCATTTC-3′	50	329	*Ava*I
6	5′-CATGAGGAAGCAGCCAGAGC-3′	5′-AGTGTCACCTGCCTCCCTGT-3′	57	333	*Hae*III
7	5′-CTGGCTAAGGTGTTCCCCTG-3′	5′-CCAGGCTCCAGAAGCTCCCA-3′	60	346	*Bam*HI
8	5′-GCTGGTGCTGTTCCCTGTCC-3′	5′-CCTCCCTTGGGCGTTTCCAG-3′	62	239	
9	5′-AGTCTGCTGTGTGTCCTG-3′	5′-CCATGCCCTGATTAAACC-3′G	50	158	
10	5′-GAGGCTGAGTGGGCTACGTC-3′	5′-AGACCTCTGTGGGGCTGGGA-3′	61	199	
11	5′-CTCTGCGGTGCCAAGCCTCA-3′	5′-TCTGTCTCCCCAGCTCGCCT-3′	62	375	*Stu*I
12	5′-GCCTTCTTCCTCCCTGTCAT-3′	5′-GAGACTCCCCAGGGCACTGT-3′	60	267	*Mae*I
13	5′-AACTTGGGGAAGGCGTGCA-3′	5′-AGAACAGGGCTGTATGGAGC-3′	57	277	*Ava*I
14	5′-AAGACCCAAGCTGCCTGACC-3′	5′-CTGGGTGCAGAGCCATATGC-3′	60	294	*Ava*II
15	5′-TGACCGCTGCTGCCTGCCAT-3′	5′-GCTTCCCAAGGACTGCCTGC-3′	61	251	
16	5′-AGGGATAGGGCCTGGGCTTC-3′	5′-TAACCTCCACCCCAAGAGAG-3′	58	192	
17	5′-AGCCACTCACTGGTCCTTCA-3′	5′-ATGGGGAGGGAATGCACACA-3′	57	240	
18	5′-CTGGCCCTGCTTGGATCATA-3′	5′-CAGCTTGTGGGAATTGGACC-3′	60	176	
19	5′-TAGTTGTGGCACATGGCTTG-3′	5′-GAGAGGAAGGATAGTGCAGA-3′	59	260	*Ava*I
20	5′-CAAAGGGAGTTTTGCCAAGG-3′	5′-GCCGGTAGACTTTCCATTCT-3′	56	311	*Eco*RV

[a] PCR was done for amplification of each of the 20 exons. The restriction enzymes indicated were used if PCR products were longer than 250 bp. Exons 1 and 4 needed 10% dimethylsulfoxide in the reaction buffer.

and patients might be associated with specific *RET* mutations. We therefore analyzed two families. In patients of one of these, MEN 2A/CLA-1, the presence of amyloid could be clearly demonstrated. Amyloid could not be demonstrated in specimens from the lesions of the patients from the other family. Because this is not a consistent feature of presumed CLA patients in previously reported MEN 2A/CLA families, however, and because all lesions were limited to the interscapular region, which is generally considered characteristic for the association of MEN 2A and CLA, family MEN 2A/CLA-2 was also included in this study (Gagel *et al*, 1989; Nunziata *et al*, 1989; Ferrer *et al*, 1991; Kousseff *et al*, 1991; Chabre *et al*, 1992; Robinson *et al*, 1992; Pacini *et al*, 1993). In the two families, we found the same *RET* mutation in codon 634 (Cys634→Arg). The mutation was present in all MEN 2A patients, some of whom also had CLA. A Cys634→Tyr mutation (G1901→A) has been reported previously in another family with MEN 2A and CLA (Ceccherini *et al*, 1994b). Although all the mutations affect codon 634, different amino acid substitutions result. The mutations found, notably Cys634→Arg, also occur frequently in MEN 2A families without CLA lesions. Although an association between MEN 2A/CLA and mutations in codon 634 may be postulated, the above-mentioned arguments make it hard to suggest a correlation between a specific *RET* mutation and the MEN 2A/CLA phenotype.

It might be suggested that the joint occurrence of MEN 2A and CLA would be due to the interaction of an apparently noncausative polymorphism and a disease-causing mutation, as has been described for the prion gene (Goldfarb *et al*, 1992). In the *RET* gene, several common noncausative polymorphisms have been found (**Table II**; Ceccherini *et al*, 1994a). None of the polymorphisms, however, seemed to co-segregate with the MEN 2A/CLA phenotype. Thus, for an explanation of the intrafamilial phenotypic variability, it may be necessary to look beyond the mutational-polymorphic genotype. Differential handling of the gene product by the paracrine growth mechanism of a particular individual may alter the pathogenesis of the condition and cause pleiotropy of the phenotype (Kousseff *et al*, 1991; Kousseff, 1992).

A search for *RET* mutations in patients from three "CLA only" families did not reveal a mutation other than already known noncausative polymorphisms. We also looked for possible co-segregation of these intragenic polymorphisms with the cutaneous phenotype in these families, but were unsuccessful. We therefore conclude that *RET* is not involved in these cases of familial CLA.

Our findings raise the question of whether the CLA found in MEN 2A and familial CLA are etiologically similar conditions. Clinically, there is a distinction in the affected sites. In MEN 2A patients, skin lesions are always found in the interscapular region, whereas in familial CLA patients, skin lesions are more generalized (Robinson *et al*, 1992). Dysfunction of the *RET* gene, which in developing mice is expressed in the peripheral nervous system (Schuchardt *et al*, 1994), might lead to pruritus and subsequently to scratching and degeneration of keratinocytes. It has been suggested that prolonged mechanical friction may produce a macular amyloidosis, or "friction amyloidosis" (Wong and Lin, 1988; Robinson *et al*, 1992). Because many chronic pruritic skin conditions do not show skin amyloidosis, however, this etiologic model might be an oversimplification.

The current results lend support to the idea that skin lesions in

Table II. Noncausative Nucleotide Changes Detected in the Coding Sequence of *RET*[a]

Exon	Nucleotide Change	Amino Acid Substitution	Restriction Site Changed[b]	Controls Tested	Allele 1 Frequency
2	GCG→GCA	Ala 45	*Eag*I (+)	52	0.71
3	GTC→GTA	Val 125	*Mbo*II (−)	49	0.98
7	GCG→GCA	Ala 432	*Bsm*I (+)	45	0.29
11	GGT→AGT	Gly 691→Ser	*Ban*I (−)	53	0.79
13	CTT→CTG	Leu 769	*Taq*I (−)	46	0.74
15	TCC→TCG	Ser 904	*Rsa*I (+)	48	0.21
18	CGC→TGC	Arg982→Cys		90	0.98

[a] Ceccherini *et al*, 1994.
[b] −, Loss of the mentioned restriction site; +, gain of the mentioned restriction site.

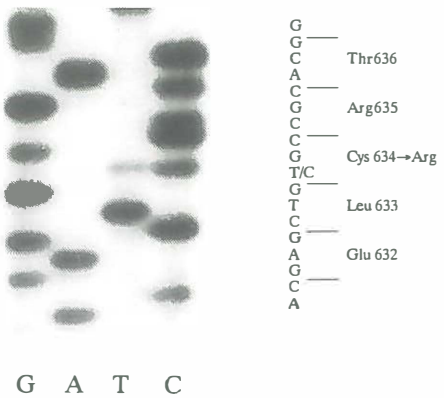

Figure 3. Partial sequence analysis of exon 11 in an affected member of family MEN 2A/CLA-1. Shown is the relevant part of the sequence containing the Cys634→Arg mutation.

familial CLA and MEN 2A/CLA patients are different from genetic, clinical, and etiologic points of view. Consequently, familial CLA patients do not appear to be at risk for MEN 2A. To settle this issue definitively, however, more data are needed. Mainly for this reason, it may still be reasonable for physicians of (apparent) familial CLA patients to have their patients screened for *RET* mutations.

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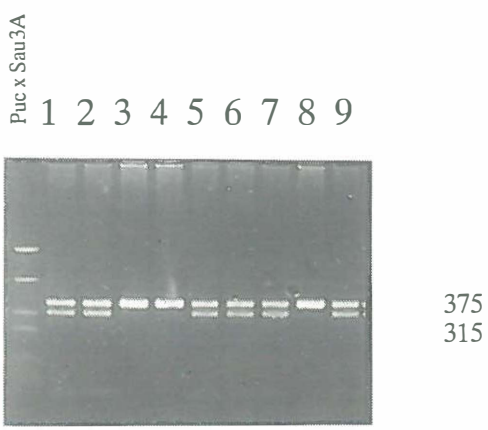

Figure 4. *Hha*I restriction patterns of exon 11 in the MEN 2A/CLA-1 family members examined. The numbers correspond to those of **Fig 2**. Affected individuals show two bands because of an extra *Hha*I site.

REFERENCES

Carlson KM, Dou S, Chi D, Scavarda N, Toshima K, Jackson CE, Wells SA, Goodfellow PJ, Donis-Keller H: Single missense mutation in the tyrosine kinase catalytic domain of the *RET* proto-oncogene is associated with multiple endocrine neoplasia type 2B. *Proc Natl Acad Sci USA* 91:1579–1583, 1994

Ceccherini I, Hofstra RMW, Yin I, Stulp RP, Barone V, Stelwagen T, Bocciardi R, Nijveen H, Bolino A, Seri M, Ronchetto P, Pasini B, Bozzano M, Buys CHCM, Romeo G: Conditions for SSCP analysis and common polymorphisms of the 20 exons of the *RET* proto-oncogene. *Oncogene* 9:3025–3029, 1994a

Ceccherini I, Romei C, Barone F, Pacini F, Martino E, Pinchera A, Romeo G: Identification of the Cys634→Tyr mutation of the *RET* proto-oncogene in a pedigree with multiple endocrine neoplasia type 2A and localized cutaneous lichen amyloidosis. *J Endocrinol Invest* 17:201–204, 1994b

Chabre O, Labat F, Pinel N, Berthod F, Tarel V, Bachelot I: Cutaneous lesion associated with multiple endocrine neoplasia type 2A: lichen amyloidosis or nostalgia paresthetica. *Henry Ford Hosp Med J* 40:245–248, 1992

Donis-Keller H, Dou S, Chi D, Carlson K, Toshima K, Lairmore TC, Howe JR, Moley JF, Goodfellow P, Wells SA: Mutations in the *RET* proto-oncogene are associated with MEN 2A and FMTC. *Hum Mol Genet* 7:851–856, 1993

Edery P, Lyonnet S, Mulligan LM, Pelet A, Dow E, Abel L, Holder S, Nihoul-Fékété C, Ponder BAJ, Munnich A: Mutations of the *RET* proto-oncogene in Hirschsprung's disease. *Nature* 367:378–380, 1994

Eng C, Smith DP, Mulligan LM, Nagai MA, Healey CS, Ponder MA, Gardner E, Scheumann FW, Jackson CE, Tunnacliffe A, Ponder BAJ: Point mutations within the tyrosine kinase domain of the *RET* proto-oncogene in multiple endocrine neoplasia type 2B and related sporadic tumours. *Hum Mol Genet* 3:237–241, 1994

Ferrer JP, Halperin I, Conget I, Alsina M, Martinez-Osaba MJ, Palou J, Bombi JA, Vilardell E: Primary localized cutaneous amyloidosis and familial thyroid carcinoma. *Clin Endocrinol (Oxf)* 34:435–439, 1991

Gagel RF, Levy ML, Donavan DT, Alford BR, Wheeler T, Tschen A: Multiple endocrine neoplasia type 2a associated with cutaneous lichen amyloidosis. *Ann Intern Med* 111:802–806, 1989

Goldfarb I, Peterson RB, Tabaton M, Brown P, LeBlanc AC, Montagna P, Cortelli P, Julien J, Vital C, Pendelbury WW, Zhaltia M, Wills PR, Hauw JJ, McKeever PE, Monari L, Schrank B, Swergold GD, Autilio-Gambetti L, Carleton Gajdusek D, Lugaresi E, Gambetti P: Fatal familial insomnia and familial Creutzfeldt-Jacob disease: disease phenotype determined by a DNA polymorphism. *Science* 258: 806–808, 1992

Hofstra RMW, Landsvater RM, Ceccherini I, Stulp RP, Stelwagen T, Luo Y, Pasini B, Höppener JWM, Ploos van Amstel HK, Romeo G, Lips CJM, Buys CHCM: A mutation in the *RET* proto-oncogene associated with multiple endocrine neoplasia type 2B and sporadic medullary thyroid carcinoma. *Nature* 367:375–376, 1994

Kousseff B: Sipple syndrome with lichen amyloidosis as a paracrinopathy. *Am J Med Genet* 42:751–753, 1992

Kousseff B, Espinoza C, Zamore GA: Sipple syndrome with lichen amyloidosis as a paracrinopathy: pleiotropy, heterogeneity, or a contiguous gene? *J Am Acad Dermatol* 25:651–657, 1991

Lips CJM, Landsvater RM, Höppener JWM, Geerdink RA, Blijman G, Jansen-Schillhorn van Veen JM, van Gills APG., de Wit M, Zewald R, Berends MJH, Beemer FA, Brouwers-Smalbraak J, Jansen RPM, Ploos van Amstel HK, van Vroonhoven TJMV, Vroom TM: Clinical screening as compared with DNA analysis in families with multiple endocrine neoplasia type 2A. *N Engl J Med* 331:823–835, 1994

Mulligan LM, Eng C, Healey CS, Clayton D, Kwok BJ, Gardner E, Ponder MA, Friling A, Jackson CE, Lehnert H, Neumann HPH, Thibodeau SN, Ponder BAJ: Specific mutations of the *RET* proto-oncogene are related to disease phenotype in MEN 2A and FMTC. *Nature Genet* 6:70–74, 1994

Mulligan LM, Kwok JBJ, Healey CS, Elsdon MJ, Eng C, Gardner E, Love DR, Mole SE, Moore J, Papi L, Ponder MA, Telenius H, Tunnacliffe A, Ponder BAJ: Germline mutations of the *RET* proto-oncogene in multiple endocrine neoplasia type 2A. *Nature* 363:458–460, 1993

Nunziata V, Giannattasio R, Di Giovanni G, D'Armiento MR, Mancini M: Hereditary localized pruritus in affected members of a kindred with multiple endocrine neoplasia type 2A (Sipple syndrome). *Clin Endocrinol (Oxf)* 30:57–63, 1989

Pacini F, Fugazzola L, Bevilacqua G, Viacava P, Nardini V, Martino E: Multiple endocrine neoplasia type 2A and cutaneous lichen amyloidosis: description of a new family. *J Endocrinol Invest* 16:295–296, 1993

Robinson MF, Furst E, Nunziata V, Brandi ML, Ferrer JP, Martins Bugalho MJG, di Giovani G, Smith RJH, Donovan DT, Alford BR, Hejtmancik JF, Colantuoni V, Quadro L, Limbert E, Halperin I, Vilardell E, Gagel RF: Characterization of the clinical features of five families with hereditary primary cutaneous lichen amyloidosis and multiple endocrine neoplasia type 2. *Henry Ford Hosp Med J* 40:249–255, 1992

Romeo G, Ronchetto P, Luo Y, Barone V, Seri M, Ceccherini I, Pasini B, Bocciardi R, Lerone M, Kääriäinen H, Martuciello G: Point mutations affecting the tyrosine kinase domain of the *RET* proto-oncogene in Hirschsprung's disease. *Nature* 367:377–378, 1994

Schuchardt A, D'Agati V, Larsson-Blomberg L, Constantini F, Pachnis V: Defects in the kidney and enteric nervous system of mice lacking the tyrosine kinase receptor Ret. *Nature* 367:380–383, 1994

Schuffenecker I, Billaud M, Calender A, Cambe B, Ginet N, Calmettes C, Modigliani E, Lenoir GM, the Groupe d'Etude des Tumeurs a Calcitonine: RET proto-oncogene mutations in French MEN 2A and FMTC families. *Hum Mol Genet* 3:1939–1943, 1994

Wong CK, Lin CS: Friction amyloidosis. *Int J Dermatol* 27:302–307, 1988

Appendix II
Oncological implications of RET gene mutations in Hirschsprung's disease

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Abstract

***Background*—Germline mutations of the RET proto-oncogene identical to those found in the tumour predisposition syndrome multiple endocrine neoplasia type 2A (MEN2A), were detected in 2.5–5% of sporadic and familial cases of Hirschsprung's disease. Some patients with Hirschsprung's disease may therefore be exposed to a highly increased risk of tumours.**

***Aims*—To define clinical use of RET gene testing in Hirschsprung's disease and related patient management from an oncological point of view.**

***Methods*—Sixty patients with Hirschsprung's disease were screened for RET mutations. In three, MEN2A type RET mutations were detected. Case reports for these three patients are presented.**

***Results and conclusions*—Only 22 families or sporadic patients with Hirschsprung's disease and MEN2A type RET mutations have been reported. Therefore, it is difficult to predict tumour risk for patients with familial or sporadic Hirschsprung's disease, and their relatives, who carry these mutations. For these mutation carriers, periodic screening for tumours as in MEN2A is advised, but prophylactic thyroidectomy is offered hesitantly. RET gene testing in familial or sporadic Hirschsprung's disease is not recommended at present outside a complete clinical research setting. In combined MEN2A/Hirschsprung's disease families RET gene testing, tumour screening, and prophylactic thyroidectomy are indicated as in MEN2A.**

(*Gut* 1998;43:542–547)

Keywords: DNA analysis; Hirschsprung's disease; multiple endocrine neoplasia type 2A; RET

Hirschsprung's disease (HD), which occurs in approximately 1 out of every 5000 newborns, is characterised by a congenital absence of enteric neurones in the distal colon and rectum.[1] Most cases are sporadic, but familial HD has been reported.[2–5] Hirschsprung's disease is associated with various chromosomal abnormalities and inherited disorders.[1 6] Two of these inherited disorders are the tumour predisposition syndromes, multiple endocrine neoplasia type 2A (MEN2A) and familial medullary thyroid cancer (FMTC). MEN2A is an autosomal dominant disorder characterised by medullary carcinoma of the thyroid (which may present as early as in the third year), phaeochromocytoma in 50% of cases, and parathyroid hyperplasia or adenoma in 20–25% of cases.[6 7] Familial medullary thyroid cancer is characterised by the familial occurrence of medullary carcinoma of the thyroid in the absence of other MEN2 tumours.

Germline mutations of the RET (REarranged during Transfection) proto-oncogene can be detected in virtually all families with MEN2A, FMTC,[8–11] and combined MEN2A/HD or FMTC/HD.[10 12–15] Germline RET mutations also occur in both familial and sporadic cases of HD without manifestations of either MEN2A or FMTC.[16–21] RET mutations were detected in 11–49% of familial HD cases and 9–35% of sporadic HD cases.[17 20 21] Different types of RET mutation can be found in these families. The mutations identical to those found in MEN2A or FMTC are of oncological interest. For the purpose of this article we will refer to these mutations as MEN2A type RET mutations. This type of RET mutation was found in 2.5–5% of the patients with HD[17 19 21–23 24] and these patients may have a high risk of developing MEN 2 tumours. Therefore, the question arises whether patients with HD and their relatives should be tested for MEN2A type RET mutations and, in case mutations are found, be screened for possible MEN 2 tumours.

While screening a series of 60 patients with HD for RET mutations,[24] we found three patients with a MEN2A type RET mutation. Using these three patients and their families as a clinical introduction to the subject, the aim of this study was to define the use of RET testing and related patient management in HD from an oncological point of view.

Case reports

PATIENT 1

This three year old girl was diagnosed with long segment HD at the age of eight days, on clinical and histological grounds. Preoperative ultrasound examination of the abdomen revealed right sided renal aplasia and during the operation a small cyst-like structure near the bifurcation of the aorta was removed. Histological examination showed renal tissue. The 31 year old mother of this patient had presented with the combination of medullary thyroid carcinoma and phaeochromocytoma at the age of 28 and been diagnosed as having MEN2A. No other cases of HD or MEN2A were diagnosed in the family (fig 1A). MEN2A might, however, have occurred in a brother who died suddenly at the age of 25 (no further

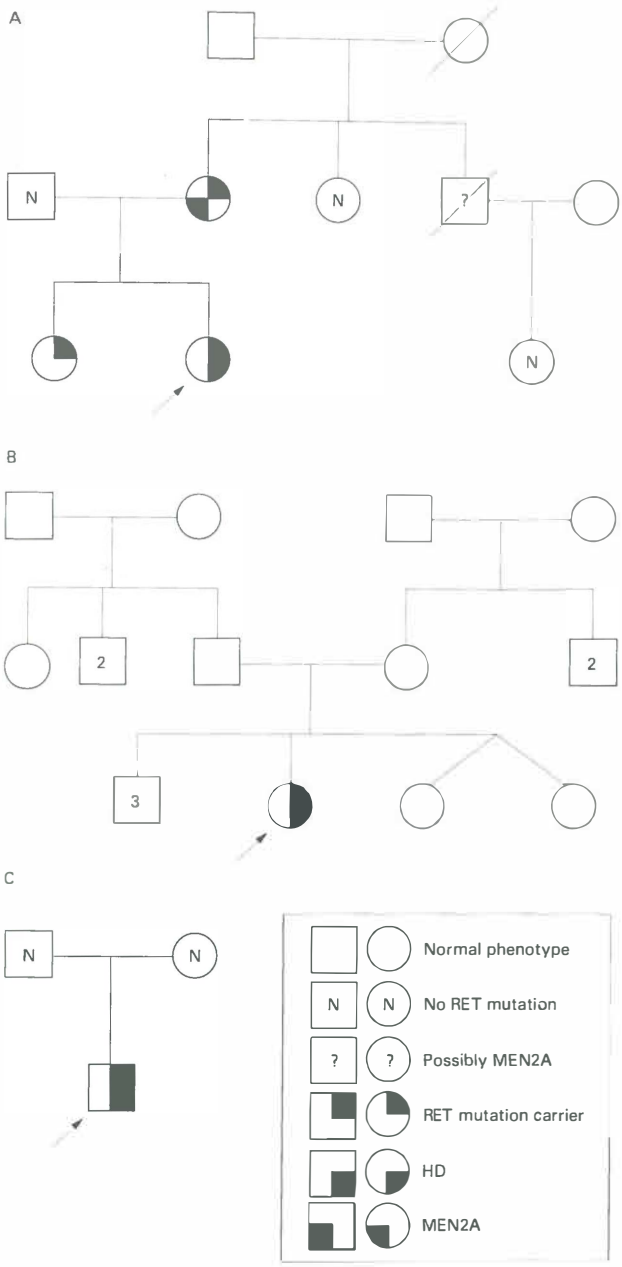

Figure 1 Pedigrees of the families of the index patients. In (A) patient 1 is indicated with an arrow. In (B) patient 2 is indicated with an arrow; she probably has combined MEN2A/HD. Her relatives could not be tested for the RET mutation or screened for MEN 2 tumours. Numbers within squares represent the additional number of asymptomatic males in that generation. In (C) patient 3 is indicated with an arrow; the mutation was not shown in his parents. HD, Hirschsprung's disease.

details available). The mother of the MEN2A patient died at the age of 55 from ovarian cancer; biochemical screening of her father at the age of 59 revealed no abnormalities. Biochemical screening of the little girl provided no evidence for C cell hyperplasia, medullary thyroid carcinoma, parathyroid involvement, or phaeochromocytoma. The RET gene was screened for mutations by means of denaturing gradient gel electrophoresis followed by direct sequencing of aberrant DNA fragments.[23 25] The girl exhibited the same germline mutation as was later found to have been previously detected in her mother in another laboratory. A TTC-TGC to TTT-CGC change was observed in codons 619–620, changing the corresponding amino acids phenylalanine-cysteine in the RET protein to phenylalanine-arginine. Cys620Arg mutations have been reported previously in MEN2A and MEN2A/HD kindreds.[10 26] Therefore, periodic screening and prophylactic thyroidectomy at the age of three to six years were recommended for this patient. The five year old sister of the HD patient was found to be an asymptomatic carrier of the RET mutation. Recently, she underwent prophylactic thyroidectomy and central node dissection; the surgical specimens revealed normal histology. Two other family members underwent DNA analysis and did not show the mutation.

PATIENT 2

This 34 year old woman was diagnosed with short segment HD at the age of eight weeks, on clinical and histological grounds. At the age of 33 she was tested for germline RET mutations (techniques as in patient 1) after an informed consent procedure including discussion of possible oncological aspects. A TGC to TAC mutation was detected in codon 609, changing its code for cysteine to one for tyrosine. Cys609Tyr mutations have previously been reported in MEN2A.[26 27] The family history was negative for HD and bowel diseases in general, and MEN 2 tumours and cancer in general (fig 1B). Relatives of the patient could not be approached for DNA analysis. After the RET mutation was found, the patient was screened biochemically for MEN 2 tumours. Her basal calcitonin level was normal: 3.88 ng/l (normal range 3.00–29.00). However, after pentagastrin stimulation, the calcitonin levels were clearly abnormal, with a peak value of 1290 ng/l (normal peak value is up to three times the normal unstimulated level) which is indicative of thyroid (C cell) pathology. Urinary screening for phaeochromocytomas was normal. The possibility of thyroid pathology was discussed with the patient and repeat pentagastrin testing (with subsequent thyroidectomy if the high calcitonin levels were confirmed) offered. However, the patient did not wish to undergo any further testing at that time because she perceived this as a direct threat to the success of a child adoption procedure she had recently started. She was offered psychosocial support.

PATIENT 3

This four year old boy was diagnosed with short segment HD at the age of six weeks, on clinical and histological grounds. Family histories of HD and congenital abnormalities in general were negative (fig 1C). A germline RET mutation was detected in this patient (techniques as in patient 1). In codon 620 the

Table 1 MEN2A type germline mutations of the RET gene found in pateints/families with Hirschsprung's disease (HD)

Mutation	*Phenotype†*	*HD/RET carriers††*	*HD + MEN2A/HD†††*	*Reference*
Cys609Trp	HD (familial)	3/7	NA	10
Cys609Tyr	HD (familial)	NR	NA	22
Cys609Tyr	HD/MEN2A	1/7	1/1	33
Cys609Tyr	HD/MEN2A	1/7	1/1	33
Cys609Tyr	HD (sporadic)	NA	NA	Current report
Cys618Arg	HD/MEN2A	2/6	2/2	13
Cys618Arg	HD/MEN2A	2/5	2/2	10
Cys618Arg	HD/FMTC	2**/3	0/3	15
Cys618Arg	HD/FMTC	2***/29	2/2***	15
Cys618Ser	HD/MEN2A	3/at least 42‡	2/3	12, 33
Cys618Ser	HD/MEN2A	4/at least 83‡	3/4	12, 33, 34
Cys618Ser	HD/MEN2A	1/3	1/1	33
Cys620Trp	HD/MEN2A	1/6	0/1	33
Cys620Arg	HD/MEN2A	2/4	1/2	33
Cys620Arg	HD/MEN2A	5/10	4/5	10
Cys620Arg	HD/MEN2A	2/2	1/2	10
Cys620Arg	HD/FMTC*	NA	NA	10
Cys620Arg	HD/MEN2A	1**/6	1/1**	14
Cys620Arg	HD/MEN2A	1/3	0/1	Current report
Cys620Arg	HD (sporadic)	NR	NA	22
Cys620Arg	HD (sporadic)	NR	NA	24
Cys620Arg§	HD (sporadic)	NA	NA	Current report

*In this family HD segregates independently of the familial RET mutation. **In an additional relative with HD RETwas not analysed. ***In three additional relatives with HD RET was not analysed.
†Phenotype refers to the classification of the family type prior to DNA analysis and (unpublished) follow up. ††HD/RET carriers refers to the number of family members diagnosed with HD out of the total number of family members with a proven RET mutation plus untested relatives with clinical MEN2A. These ratios are given mainly to show the incomplete penetrance of the mutations with regard to the HD phenotype, not to provide a precise figure. The ratios given are probably an overestimation of penetrance as not all unaffected relatives have been tested for RET mutations; uncertain cases were not included. †††HD + MEN2A/HD refers to the number of patients with both HD and MEN2A out of the total number of relatives with HD. Young relatives with HD may of course develop MEN2A tumours at a later age; uncertain cases were not included.
‡Exact numbers not published.
§De novo mutation.
NA, not applicable; NR, not reported.
The International RET Mutation Consortium recently published an update of their dataset [3] and report on five MEN2A/HD families and one FMTC/HD family with a Cys620Arg mutation in 5/6 cases and in the remaining case a Cys618Arg germline mutation of RET. It cannot be deduced from that report whether those families are identical to the families described in the reports listed in this table.

same TGC to CGC change as in patient 1 was observed, changing the corresponding amino acid in the RET protein from cysteine to arginine. This mutation was not detected in the peripheral blood lymphocytes of his parents, which implies that it is a de novo (spontaneous) mutation; however, germline mosaicism in one of the parents (presence of the mutation in only part of the germ cells), a phenomenon which has been reported in RET gene associated HD,[21] cannot be excluded. Periodic screening of the boy, but not prophylactic thyroidectomy, has been recommended.

Discussion

Given our present knowledge, we believe that patients with familial or sporadic HD, and their relatives, should only be tested for RET gene mutations in a complete clinical research setting. If MEN2A type RET mutations are found, then they should be screened for MEN 2 tumours. We considered the following.

Firstly, the molecular background should be taken into account. The RET proto-oncogene encodes a transmembrane receptor, which has glial cell line derived neurotrophic factor (GDNF) and neurturin (NTN) as its ligands.[28 31] This receptor is believed to transduce signals involved in cell growth and in differentiation of neural crest cell derived tissues, including thyroid C cells (involved in medullary thyroid carcinoma), the adrenal medulla (involved in phaeochromocytoma), enteric neurones, and also the parathyroid glands and kidneys which are not neural crest cell derived.

Different types of mutation of RET can have a strikingly different impact on the function of the receptor it codes for. On the one hand, mutations in MEN 2 and FMTC are presumably or have been proved to be of the activating type, whereby ligand binding is no longer necessary for substrate activation (phosphorylation).[15] Activating mutations are thought to predispose to tumour development as they provide the cells with a proliferative advantage. The RET mutations found in all combined MEN2A (or FMTC)/HD families and in 2.5–5% of sporadic and familial HD cases analysed to date are of this type[10 13 21 22] (table 1).

On the other hand, 95–97.5% of the mutations found in HD are presumably or have been proved to be of the inactivating type, which is not expected to predispose to tumour development.[15 38] Instead, these mutations are thought to cause defective migration, proliferation, differentiation, or survival of the enteric neuroblasts which form the enteric ganglia.[39] This hypothesis of a loss of RET function leading to HD is supported by the absence of enteric ganglia in so called knockout mice which lack both copies of the RET gene.[40] These mice also show renal agenesis or severe renal dysplasia.[40] Patient 1 is one of the few clinical examples of a proved (heterozygous) RET mutation associated with both HD and renal dysplasia. Similar cases have been reported by Attie and colleagues[21] and McGaughran *et al*,[41] respectively.

The occurrence of HD, generally associated with lack of RET function, in the presence of presumed activating tumour predisposing RET mutations seems to be a contradiction. Another puzzling finding is that, although in the majority

of MEN2A families RET mutations affect cysteine codon 634,[9] no such mutations have been found in MEN2A/HD families (table 1). Mutations of three other cysteine codons, namely 609, 618, and 620, account for all cases reported to date.

Recent findings by Ito and colleagues[42] may provide a clue to these apparent oddities. These authors showed that mutations of RET in cysteine codons 609, 618, and 620 were associated with a much lower expression of the RET protein on the cell surface than mutations of cysteine in codon 634. This is important because, in order to act properly, the receptor needs to be positioned within the cell membrane. One could therefore speculate that the decreased number of receptors at the cell surface falls below the critical threshold needed for successful development of the enteric ganglia. In the C cells, adrenal medulla, and parathyroid gland, however, intrinsic activation of the mutated receptor (though present in lower numbers) may lead to tumour formation. Ito *et al* speculate that the lower number of receptors at the cell surface will cause tumours which may differ in clinical behaviour from those seen in patients with codon 634 mutations,[12] but this hypothesis has yet to be verified.

Secondly, in addition to the molecular data, the predictive clinical value of finding RET mutations in patients with HD and their relatives has to be considered. Given a specific germline MEN2A type RET mutation, it is not yet possible to make an exact prediction of the actual risks of developing an MEN 2 tumour and HD. MEN2A, MEN2A/HD, and HD families may show explicit intrafamilial and interfamilial differences in the expression of their mutant genes. These differences relate to the age of onset of possible tumours, the presence or absence of phaeochromocytomas, and possible parathyroid involvement (table 1). Similar variation is seen with regard to HD: both short and long segment HD as well as the absence of HD may be associated with the same RET mutation.[10 12 21 33 43] This seriously limits the use of RET testing to support counselling for HD risks in offspring.

The phenotypic expression of germline RET mutations can apparently be influenced by additional genetic factors (modifying genes) or environmental factors. With regard to MEN2A, there are enough data available to make general estimates of tumour risks. In contrast, the total number of index patients with HD with MEN2A type mutations, observed to date is very small. Including our own three patients, 22 cases have been reported so far and only five of these belong to the most difficult category for risk estimation: those with no family history of MEN 2 tumours.

Although family history might be an indication of tumour risk, unfortunately, a negative family history of MEN 2 tumours for patients with HD and MEN2A type RET mutations does not mean that the tumour risk is not increased. Families may be small and MEN 2 tumours may be asymptomatic. After biochemical screening for tumours and pedigree expansion and verification, at least some sporadic or familial patients with HD with MEN2A type mutations might actually turn out to be MEN2A/HD patients or families. Our patient 2 is a likely candidate for the combined MEN2A/HD disorder and it could be that other family members are at risk for MEN 2 tumours and should, if circumstances allow, be tested. In cases of de novo RET mutations, as in our patient 3, there is of course no way to deduce tumour risk from family history. In many sporadic or familial HD only cases it will be impossible to predict whether or not tumour risk is increased as substantially as it is in MEN2A. Only in exceptional cases, for example, the presence of many—especially older—relatives with a MEN 2 type mutation and a negative family history of MEN 2 tumours, do the data suggest that that particular family is not exposed to a substantially increased risk of these tumours.

Predicting tumour risk for RET mutation carriers from MEN2A (or FMTC)/HD families, such as patient 1, seems less difficult. The published cases indeed suggest that the risk is as high as for MEN2A (or FMTC) only families. This also applies to patients with HD in these families, as the large majority of family members with HD also developed MEN 2 tumours (table 1). Furthermore, some of the patients with HD without MEN 2 tumours are still young and may develop these tumours later in life.

Thirdly, the options for tumour prevention or early intervention should be taken into account. The prognosis for patients with MEN 2 tumours detected at the symptomatic stage is worse compared with that for patients with tumours detected by screening.[44 45] Therefore, although the risk of a tumour is not yet known, we think that patients and relatives of sporadic or familial HD families known to carry a MEN 2 type RET mutation should undergo biochemical screening as in MEN2A. Screening, which starts at the age of three to five years, includes a yearly basal calcitonin test, a calcitonin test under pentagastrin stimulation to detect C cell hyperplasia/medullary thyroid carcinoma, measurement of serum calcium levels to test for parathyroid hyperplasia/adenomas, and measurement of catecholamines in urine to detect phaeochromocytomas.[27 46–48]

As in MEN2A, one should be aware of the possibility that (moderately) raised calcitonin levels may simply reflect MEN 2 independent C cell hyperplasia, which is not a precursor of medullary thyroid carcinoma. This hyperplasia is found in approximately 5% of the general population.[49] A false positive response to pentagastrin stimulation in RET mutation negative relatives has also been reported when C cell hyperplasia was not shown.[50] The test for calcitonin levels may also give false negative results. In some MEN2A/FMTC families medullary thyroid carcinoma has been encountered in children with normal pentagastrin stimulated calcitonin levels who underwent thyroidectomy after DNA diagnosis.[48 51 52] For

this reason prophylactic thyroidectomy is performed as early as the age of five years in confirmed RET mutation carriers in MEN2A or FMTC families with normal (stimulated) plasma calcitonin levels, although some clinicians prefer to wait until the pentagastrin test results are abnormal.[47 48 52] As it is difficult to predict the actual tumour risk for patients with sporadic or familial HD associated with a MEN2A type RET mutation, we are hesitant to suggest prophylactic thyroidectomy (and central lymph node dissection) when calcitonin levels are normal.

Long term follow up of patients and families with HD with different types of RET mutations is needed to evaluate the clinical value of testing for the RET gene in HD. In the meantime, we propose, based on our review, that such testing should be limited to a complete clinical research setting in which clinical genetic assessment, screening, and treatment for MEN 2 tumours are available, and long term follow up of the families is carefully registered. In contrast, RET testing and clinical management in HD/MEN2A (or FMTC) families should follow the guidelines for MEN2A. In all cases, informed consent, including a discussion of possible consequences of DNA testing, should be obtained prior to testing.

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1 Dehner LP. *Pediatric surgical pathology*. Baltimore: Williams & Wilkins, 1987:394–7.
2 Passarge E. The genetics of Hirschsprung's disease: evidence for heterogeneous etiology and a study of sixty-three families. *N Engl J Med* 1967;**276**:138–43.
3 Badner JA, Sieber WK, Garver KL, *et al*. A genetic study of Hirschsprung disease. *Am J Hum Genet* 1990;**46**:568–80.
4 McKusick VA. *Mendelian inheritance in man: a catalog of human genes and genetic disorders*. Baltimore: The Johns Hopkins University Press, 1994.
5 Schinzel A. *Human cytogenetics database*. Oxford: Oxford University Press, 1994.
6 Ponder BAJ, Ponder MA, Coffey R, *et al*. Risk estimation and screening in families of patients with medullary thyroid carcinoma. *Lancet* 1988;i:397–401.
7 Hodgson SV, Maher ER. *A practical guide to human cancer genetics*. Cambridge: Cambridge University Press, 1993.
8 Mulligan LM, Marsh DJ, Robinson BG, *et al*. Genotype phenotype correlation in multiple endocrine neoplasia type 2: report of the International RET Mutation Consortium. *J Intern Med* 1995;**238**:343–6.
9 Eng C, Clayton D, Schuffenecker I, *et al*. The relationship between specific RET proto-oncogene mutations and disease phenotype in multiple endocrine neoplasia type 2: International RET Mutation Consortium analysis. *JAMA* 1996;**276**:1575–9.
10 Mulligan LM, Eng C, Attie T, *et al*. Diverse phenotypes associated with exon 10 mutations of the RET proto-oncogene. *Hum Mol Genet* 1994;**3**:2163–7.
11 Smith DP, Eng C, Ponder BAJ. Mutations of the RET proto-oncogene in the multiple endocrine neoplasia type 2 syndromes and Hirschsprung disease. *J Cell Sci* 1994;**18**: 43–9.
12 Borst MJ, Vancamp JM, Peacock ML, *et al*. Mutational analysis of multiple endocrine neoplasia type 2A associated with Hirschsprung's disease. *Surgery* 1995;**117**:386–91.
13 Caron P, Attie T, David D, *et al*. C 618 R mutation in exon 10 of the RET proto-oncogene in a kindred with multiple endocrine neoplasia type 2A and Hirschsprung's disease — Comments. *J Clin Endocrinol Metab* 1996;**81**:2731–3.
14 Blank RD, Sklar CA, Dimich A, *et al*. Clinical presentations and RET protooncogene mutations in seven multiple endocrine neoplasia type 2 kindreds. *Cancer* 1996;**78**: 1996–2003.
15 Peretz H, Luboshitsky R, Baron E, *et al*. Cys 618 Arg mutation in the RET proto-oncogene associated with familial medullary thyroid carcinoma and maternally transmitted Hirschsprung's disease suggesting a role for imprinting. *Hum Mutat* 1997;**10**:155–9.
16 Pelet A, Attie T, Goulet O, *et al*. De-novo mutations of the RET proto-oncogene in Hirschsprung's disease. *Lancet* 1994;**344**:1769–70.
17 Yin L, Barone V, Seri M, *et al*. Heterogeneity and low detection rate of RET mutations in Hirschsprung disease. *Eur J Hum Genet* 1994;**2**:272–80.
18 Romeo G, Ronchetto P, Luo Y, *et al*. Point mutations affecting the tyrosine kinase domain of the RET proto-oncogene in Hirschsprung's disease. *Nature* 1994;**367**:377–8.
19 Attie T, Pelet A, Sarda P, *et al*. A 7 bp deletion of the RET proto-oncogene in familial Hirschsprung's disease. *Hum Mol Genet* 1994;**3**:1439–40.
20 Edery P, Lyonnet S, Mulligan LM, *et al*. Mutations of the RET proto-oncogene in Hirschsprung's disease. *Nature* 1994;**367**:378–80.
21 Attie T, Pelet A, Edery P, *et al*. Diversity of RET proto-oncogene mutations in familial and sporadic Hirschsprung disease. *Hum Mol Genet* 1995;**4**:1381–6.
22 Angrist M, Bolk S, Thiel B, *et al*. Mutation analysis of the RET receptor tyrosine kinase in Hirschsprung disease. *Hum Mol Genet* 1995;**4**:821–30.
23 Hofstra RMW, Osinga J, Stulp RP, *et al*. Mutations in three genes are found associated with the development of Hirschsprung disease: RET, EDNRB and EDN2. *Am J Hum Genet* 1996;**59**(suppl):A263.
24 Seri M, Yin L, Barone V, *et al*. Frequency of RET mutations in long- and short-segment Hirschsprung disease. *Hum Mutat* 1997;**9**:243–9.
25 Hofstra RMW, Sijmons RH, Stelwagen T, *et al*. RET mutation screening in familial cutaneous lichen amyloidosis and in skin amyloidosis associated with multiple endocrine neoplasia. *J Invest Dermatol* 1996;**107**:215–18.
26 Donis-Keller H. The RET proto-oncogene and cancer. *J Intern Med* 1995;**238**:319–25.
27 Gagel RF, Cote GJ, Bugalho MJGM, *et al*. Clinical use of molecular information in the management of multiple endocrine neoplasia type 2A. *J Intern Med* 1995;**238**:333–41.
28 Trupp M, Arenas E, Fainzilber M, *et al*. Functional receptor for GDNF encoded by the c-ret proto-oncogene. *Nature* 1996;**381**:785–8.
29 Durbec P, Marcos-Gutierrez CV, Kilkenny C, *et al*. GDNF signalling through the Ret receptor tyrosine kinase. *Nature* 1996;**381**:789–93.
30 Buj-Bello A, Adu J, Pinon LGP, *et al*. Neurturin responsiveness requires a GPI-linked receptor and the Ret receptor tyrosine kinase. *Nature* 1997;**387**:721–4.
31 Klein RD, Sherman D, Ho WH, *et al*. A GPI-linked protein that interacts with Ret to form a candidate neurturin receptor. *Nature* 1997;**387**:717–21.
32 Santoro M, Carlomagno F, Romano A, *et al*. Activations of RET as a dominant transforming gene by germline mutations of MEN2A and MEN2B. *Science* 1995;**267**:381–3.
33 Decker RA, Peacock ML, Watson P. Hirschsprung disease in MEN2A: increased spectrum of RET exon 10 genotypes and strong genotype-phenotype correlation. *Hum Mol Genet* 1998;**7**:129–34.
34 Decker RA. Long-term follow-up of a large North American kindred with multiple endocrine neoplasia type 2A. *Surgery* 1992;**112**:1066–72.
35 Pasini B, Borrello MG, Greco A, *et al*. Loss of function effect of RET mutations causing Hirschsprung disease. *Nat Genet* 1995;**10**:35–40.
36 Cosma MP, Panariello L, Quadro L, *et al*. A mutation in the RET proto-oncogene in Hirschsprung's disease affects the tyrosine kinase activity associated with multiple endocrine neoplasia type 2A and 2B. *Biochem J* 1996;**314**:397–400.
37 Carlomagno F, de Vita G, Berlingieri MT, *et al*. Molecular heterogeneity of RET loss of function in Hirschsprung's disease. *EMBO J* 1996;**15**:2717–25.
38 Iwashita T, Murakami H, Asai N, *et al*. Mechanism of Ret dysfunction by Hirschsprung mutations affecting its extracellular domain. *Hum Mol Genet* 1996;**5**:1577–80.
39 Kedinger M, Newgreen D. The gut and enteric nervous system. In: Thorogood P, ed. *Embryos, genes, and birth defects*. Chichester: John Wiley & Sons Ltd, 1997:153–96.
40 Schuchardt A, D'Agati V, Larsson-Blomberg L, *et al*. Defects in the kidney and enteric nervous system of mice lacking the tyrosine kinase receptor RET. *Nature* 1994;**367**: 380–3.
41 McGaughran J, Read AP, Luo Y, *et al*. Hirschsprung's disease and unilateral renal agenesis in a child with a RET-protooncogene abnormality. *Eur J Hum Genet* 1996;**4**:69.
42 Ito S, Iwashita T, Asai N, *et al*. Biological properties of ret with cysteine mutations correlate with multiple endocrine neoplasia type 2A, familial medullary thyroid carcinoma, and Hirschsprung's disease phenotype. *Cancer Res* 1997; **57**:2870–2.
43 Lyonnet S, Bolino A, Pelet A, *et al*. A gene for Hirschsprung disease maps to the proximal long arm of chromosome 10. *Nat Genet* 1993;**4**:346–50.
44 Gagel RF, Tashjian AH Jr, Cummings T, *et al*. The clinical outcome of prospective screening for multiple endocrine neoplasia type 2a. An 18-year experience. *N Engl J Med* 1988;**318**:478–84.
45 Bergholm U, Bergstrom R, Ekbom A. Long term follow-up of patients with medullary carcinoma of the thyroid. *Cancer* 1997;**79**:132–8.
46 Calmettes C, Ponder BAJ, Fischer JA, *et al*. Early diagnosis of the multiple endocrine neoplasia type 2 syndrome: consensus statement. *Eur J Clin Invest* 1992;**22**:755–60.
47 Ledger GA, Khosla S, Lindor NM, *et al*. Genetic testing in the diagnosis and management of multiple endocrine neoplasia type II. *Ann Intern Med* 1995;**122**:118–24.

48 Wohllk N, Cote GJ, Evans DB, *et al*. Application of genetic screening information to the management of medullary thyroid carcinoma and multiple endocrine neoplasia type 2. *Endocrinol Metab Clin North Am* 1996;25:1–25.
49 Landsvater RM, Rombouts AG, te Meerman GJ, *et al*. The clinical implications of a positive calcitonin test for C-cell hyperplasia in genetically unaffected members of an MEN2A kindred. *Am J Hum Genet* 1993;52:335–42.
50 Marsh DJ, McDowall D, Hyland VJ, *et al*. The identification of false positive responses to the pentagastrin stimulation test in RET mutation negative members of MEN2A families. *Clin Endocrinol* 1996;44:213–20.
51 Lips CJM, Landsvater RM, Hoppener JWM, *et al*. Clinical screening as compared with DNA analysis in families with multiple endocrine neoplasia type 2A. *N Engl J Med* 1994; 331:828–35.
52 Wells SA Jr, Chi DD, Toshima K, *et al*. Predictive DNA testing and prophylactic thyroidectomy in patients at risk for multiple endocrine neoplasia type 2A. *Ann Surg* 1994;220: 237–50.

Appendix III
Urinary tract cancer and hereditary non-polyposis colorectal cancer: risks and screening options

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ABSTRACT

Purpose: We investigate the risk of the different types of urinary tract cancer in hereditary nonpolyposis colorectal cancer families and review screening options.

Materials and Methods: We retrospectively calculated the relative and cumulative risks of developing urinary tract cancer by comparing tumor occurrence in patients and their first degree relatives in the Dutch hereditary nonpolyposis colorectal cancer registry with those in the general Dutch population. A person-year analysis was used, including data on 1,321 individuals from 50 hereditary nonpolyposis colorectal cancer families.

Results: The relative risk of developing transitional cell cancer of the renal pelvis or ureter was 14.04 (95% confidence interval 6.69 to 29.45, $p < 0.05$) and the cumulative risk was 2.6%. The risks of renal (excluding renal pelvis) and bladder cancers were not significantly increased. Urinary tract cancer was diagnosed at a relatively young age and many women were affected. Some familial clustering was observed.

Conclusions: Our findings indicate that hereditary nonpolyposis colorectal cancer is associated with an increased risk of transitional cell cancer of the upper urinary tract. The cumulative risk is relatively low, although a subset of hereditary nonpolyposis colorectal cancer families may be exposed to a much higher risk. As yet nothing is known of the clinical impact of screening for urinary tract cancer in cases of hereditary nonpolyposis colorectal cancer. In a research setting screening by excretory urography of hereditary nonpolyposis colorectal cancer families with a strong history of upper urinary tract cancer should be considered.

KEY WORDS: neoplastic syndromes, hereditary; colorectal neoplasms, hereditary, nonpolyposis; urinary tract

Some tumors of the urinary tract develop as a manifestation of a hereditary cancer predisposition syndrome,[1,2] of which the most frequent is hereditary nonpolyposis colorectal cancer (the Lynch syndrome).[3] Hereditary nonpolyposis colorectal cancer is an autosomal dominant disorder characterized by a 70 to 90% lifetime risk of colorectal cancer and an excess of extracolonic tumors, including cancer of the endometrium (30 to 50% risk), ovaries, stomach and urinary tract.[3-5]

Hereditary nonpolyposis colorectal cancer families benefit from periodic screening for colorectal cancer.[6,7] Reduction of colorectal cancer morbidity and mortality in these families will make prevention and early detection of the other tumors associated with it, including those of the urinary tract, more important. We questioned which types of urinary tract cancer belong to the hereditary nonpolyposis colorectal cancer tumor spectrum and whether screening for urinary tract cancer should be offered to affected families. We investigated the hereditary nonpolyposis colorectal cancer associated risk for urinary tract cancer and reviewed the literature on available tumor screening techniques. We calculated relative and cumulative risks of developing different types of urinary tract cancer by retrospectively comparing patients and their first degree relatives in the Dutch hereditary nonpolyposis colorectal cancer registry with the general Dutch population.

MATERIAL AND METHODS

The data of 1,321 hereditary nonpolyposis colorectal cancer patients and first degree relatives (684 men, 631 women, 6 unknown gender) from 50 families listed in the Dutch registry were analyzed. Colorectal cancer had been diagnosed in 312 of these subjects. All families met the minimal criteria for hereditary nonpolyposis colorectal cancer, known as the Amsterdam criteria,[3] which include at least 3 relatives (1 a first degree relative of the other 2) with histologically verified colorectal cancer, colorectal cancer involving at least 2 successive generations, at least 1 case of colorectal cancer diagnosed before age 50 and exclusion of familial adenomatous polyposis. In some patients the diagnosis of cancer was based on data from hospital records instead of the original pathology reports when they were not available. None of the registered families was ascertained through a proband with urinary tract cancer. Information was collected on all subjects through personal interviews and review of medical charts.

The occurrence of urinary tract cancer from birth until death or loss to followup was counted in all subjects. Observation time was until date of diagnosis of urinary tract cancer, death, date of last contact or closing date of the study (January 1, 1995), whichever came first. Person-years at risk (total 60,237) were stratified using gender and age specific categories with 5-year intervals up to 85 years. The relative risk was defined as the ratio between the observed and expected number of tumors. The 95% confidence intervals (CI) were calculated assuming a Poisson distribution. The expected numbers of urinary tract cancer were calculated by multiplying the person-years in the different gender and age specific categories by gender and age specific incidence rates obtained from the population based Dutch National Cancer Registry of 1990.[8,9] In our calculations we combined renal pelvic and ureteral tumors because the Dutch National Cancer Registry only provides combined incidence rates for these tumors.

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RESULTS

A total of 17 urinary tract cancers were observed (see table). Two patients had multiple primary urinary tract cancers and some familial clustering was also observed. In 1 family 4 (see figure) and in 2 families 2 relatives had urinary tract cancer.

Renal cancer, excluding the renal pelvis, was observed in 5 patients with a mean age at diagnosis of 62 years (range 52 to 70). Relative risk was 2.04 (95% CI 0.85 to 4.89, $p > 0.05$). In 2 of the 5 patients with renal cancer other primary malignancies were present, which were typical of hereditary nonpolyposis colorectal cancer. Renal pelvic or ureteral cancer was detected in 7 patients with a mean age at diagnosis of 58 years (range 44 to 70). Relative risk was 14.04 (95% CI 6.69 to 29.45, $p < 0.05$). Cumulative lifetime risk was estimated at 2.6%, whereas this risk was 0.25% (men) and 0.10% (women) in the general Dutch population. Multiple primary tumors in 5 of 6 patients were typical of hereditary nonpolyposis colorectal cancer. Bladder cancer was observed in 5 patients with a mean age at diagnosis of 60 years (range 51 to 76). Relative risk was 1.52 (95% CI 0.63 to 3.66, $p > 0.05$). In 4 patients multiple primary tumors were present, which were typical of hereditary nonpolyposis colorectal cancer in 2.

DISCUSSION

Our results indicate an increased risk of transitional cell cancer of the renal pelvis and ureter in cases of hereditary nonpolyposis colorectal cancer. Since sporadic transitional cell cancers of the renal pelvis, ureter and bladder are commonly thought to share risk factors, it may seem unusual that bladder cancer risk does not appear to be increased in cases of hereditary nonpolyposis colorectal cancer, while cancer risk at the other 2 sites does. However, these 3 sites may differ in susceptibility to specific risk factors[10] and the oncogenetic pathways in the setting of hereditary nonpolyposis colorectal cancer may differ from those in sporadic cases. We compared our results with those of Watson and Lynch,[5] and Aarnio[11] and Vasen[12] et al, who had previously reported on risks of urinary tract cancer in cases of hereditary nonpolyposis colorectal cancer. However, their studies differed from ours in the categories of tumors examined, type of risk that was calculated (relative versus cumulative risk) and type of population (proved or putative gene carriers versus patients and their first degree relatives).

Watson and Lynch calculated the relative risk of kidney cancer, including renal cell cancer and transitional cell cancer of the renal pelvis, and transitional cell cancer of the ureter and bladder in a data set of more than 1,300 members (patients and their first degree relatives) from 23 hereditary nonpolyposis colorectal cancer kindreds.[5] They reported a relative risk of 3.2 ($p < 0.01$) for kidney cancer with a mean patient age at diagnosis of 66 years and 22.0 ($p < 0.001$) for ureteral cancer with a mean age at diagnosis of 56 years. Bladder cancer risk was not increased. As in our study the highest risks were associated with transitional cell cancer of the upper urinary tract.

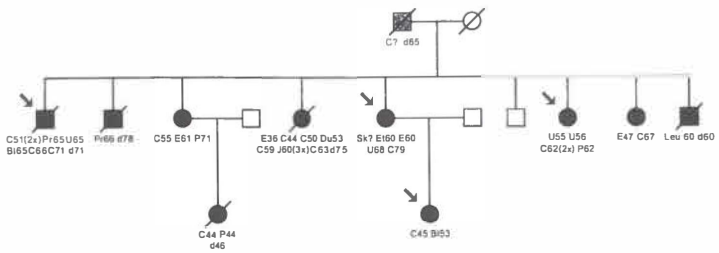
Aarnio et al calculated cumulative lifetime risks for urinary tract cancer, which they did not further specify, in 293 putative hereditary nonpolyposis colorectal cancer gene carriers included in the Finnish registry.[11] The risk for urinary tract cancer was 0.8% at age 50 and 10.2% at age 80 years. As in our study these data indicate that the cumulative risk for developing urinary tract cancer in cases of hereditary nonpolyposis colorectal cancer is low compared with the risks of developing colorectal or endometrial cancer.

Since approximately 50% of the first degree relatives of our patients did not carry the mutated hereditary nonpolyposis colorectal cancer gene, our findings in them and even the patients (some may have had sporadic colorectal cancer and would then have been misclassified) are likely to underestimate the cancer risk in cases of hereditary nonpolyposis colorectal cancer. This view is supported by the study of Vasen et al.[12] They studied a group of 210 proved gene carriers from 19 hereditary nonpolyposis colorectal cancer families, and calculated a relative risk of 75.3 (95% CI 31.3 to 180.9, $p < 0.05$), which is much higher than what we found, for developing cancer of the combined kidney and renal pelvis/ureter in carriers of the hMSH2 mutations. Risk of developing these tumors was not increased in carriers of hMLH1 mutations. Neither hMLH1 nor hMSH2 mutation carriers had an increased risk of bladder cancer.

We observed a shift towards an early age of onset of the tumors, especially for renal pelvis/ureteral cancer, and a relatively large number of affected women. The male-to-female ratio was 0.9 for urinary tract cancer in general and a ratio of 0.4 was noted for renal pelvis/ureteral cancer in particular, whereas in the general Dutch population men were affected more frequently than women. Similar observations were reported by the International Collaborative Group on hereditary nonpolyposis colorectal cancer, which collected data on 58 patients with urinary tract cancer from 8 countries.[13] The results indicated that transitional cell cancer is the most frequent tumor type, men and women are affected equally, urinary tract cancer develops at an earlier age than

Urinary tract cancer in Dutch hereditary nonpolyposis colorectal cancer patients and their first degree relatives

Family No. — Sex	Tumor	Histology	Age at Diagnosis	Additional Tumors (age at diagnosis)
1 — M	Pelvis/ureter, bladder	Transitional cell Ca	65	Colorectal ×4 (51, 66, 71), prostate (65)
1 — F	Pelvis/ureter	Transitional cell Ca	68	Skin(?), ethmoid bone (60), endometrium (60), colorectal (79)
1 — F	Pelvis/ureter	Transitional cell Ca	55	Colorectal ×2 (62)
1 — F	Bladder	Transitional cell Ca	53	Colorectal (45)
5 — M	Kidney	Renal cell Ca	62	
13 — M	Kidney	Clinical diagnosis (including radiological studies), no histology	70	Colorectal ×5 (62, 70)
19 — F	Kidney	Clinical diagnosis (including radiological studies), no histology	61	Breast (51)
19 — M	Bladder	Clinical diagnosis (including radiological studies), no histology	51	Colorectal (51)
23 — F	Pelvis/ureter	Transitional cell Ca	51	Colorectal (?)
34 — M	Bladder	Transitional cell Ca	76	
36 — F	Pelvis/ureter	Transitional cell Ca	44	
36 — F	Pelvis/ureter, bladder	Transitional cell Ca, Transitional cell Ca	53, 54	
40 — F	Kidney	Clinical diagnosis (including radiological studies), no histology	65	
51 — M	Kidney	Clinical diagnosis (including radiological studies), no histology	52	
74 — M	Pelvis/ureter	Transitional cell Ca	70	Colorectal (55)



Part of hereditary nonpolyposis colorectal cancer family 1. All relatives with cancer are shown, unaffected relatives in younger generations have been omitted for clarity. Solid black squares and circles reflect men and women with histologically verified diagnosis. Gray square represents man with diagnosis based on family history only. All relatives with urinary tract cancer are indicated with an arrow. *C*, colorectal cancer; *Bl*, bladder cancer; *d*, deceased; *Du*, duodenal cancer; *E*, endometrial cancer; *Et*, ethmoid bone tumor; *J*, jejunum cancer; *Leu*, leukemia; *P*, benign colorectal polyp; *Pr*, prostate cancer; *Sk*, nonmelanoma skin cancer; *U*, transitional cell cancer of renal pelvis or ureter; Numbers after diagnostic code refer to age at diagnosis, question mark indicates age unknown and number after *d* is age at death.

sporadic urinary tract cancer and a large proportion of patients with urinary tract cancer have a second primary tumor (72% in their data set compared with 60% in our study).

In addition to evidence from epidemiological studies, one could look for molecular findings in support of a causal relationship between hereditary nonpolyposis colorectal cancer and the different types of urinary tract cancer. Hereditary nonpolyposis colorectal cancer is characterized by germline mutations in a number of genes responsible for deoxyribonucleic acid mismatch repair, including hMLH1, hMSH2, hPMS1, hPMS2 and hMSH6 (or GTBP).[14,15] When both copies of a hereditary nonpolyposis colorectal cancer gene have been mutated, 1 in the germline and 1 during the course life (referred to as a somatic mutation), mismatch repair will be deficient, causing genomic instability, typically of the deoxyribonucleic acid stretches with repetitive sequences known as the microsatellites. Genomic instability may subsequently lead to tumor development.[16] Therefore, these tumors are characterized by a somatic mutation, often a loss (loss of heterozygosity) of the normal (wild type) copy of the hereditary nonpolyposis colorectal cancer gene as well as microsatellite instability.[17] The presence of microsatellite instability alone is not evidence of a causal relationship between tumor development and hereditary nonpolyposis colorectal cancer, because it frequently occurs in sporadic cancer as well, including that of the urinary tract.[18–23]

Unfortunately, only limited molecular data on urinary tract cancer in patients with hereditary nonpolyposis colorectal cancer, or its variant the Muir-Torre syndrome,[24] have been reported. Only 3 tumors were studied for microsatellite instability[25,26] and none was studied for loss of heterozygosity. Additional molecular studies are clearly needed to gain insight in the molecular events leading to the development of urinary tract tumors in cases of hereditary nonpolyposis colorectal cancer. These insights might be of help in developing strategies for tumor prevention.

Screening for kidney tumors is part of the preventive programs in patients and their first degree relatives from families with von Hippel-Lindau disease[27] and hereditary papillary renal cancer.[28,29] Compared with these disorders, the risk of urinary tract cancer in hereditary nonpolyposis colorectal cancer is low. Therefore, it is questionable whether screening for these tumors in all families is justified, even if the techniques are sensitive and specific. However, some families may be exposed to much higher risks for urinary tract cancer than those calculated in the total group. Indeed, a significant difference among families has been observed in the frequency of upper urological tract cancer,[5] possibly and partly related to the type of hereditary nonpolyposis colorectal cancer genes mutated.[12] Therefore, a family history of urinary tract cancer might be an indicator of this higher than average risk and be used to select families for urinary tract cancer screening. Unfortunately, especially smaller hereditary nonpolyposis colorectal cancer families need not to have manifested the increased risk for these tumors. Thus, one could choose to include also proved carriers of the gene mutations in the screening program to avoid having to screen the large (50%) group of first degree relatives without hereditary nonpolyposis colorectal cancer germline mutations. However, in approximately half of the families that meet the clinical (Amsterdam) criteria, germline hereditary nonpolyposis colorectal cancer gene mutations have not yet been identified.

Tumor risk is but 1 factor of deciding whether to screen. Tumor morbidity and mortality, sensitivity and specificity of screening techniques, benefits from early detection, physical burden of screening and economical issues should be considered as well. Since prognosis of early stage urinary tract cancer is better than that of more advanced disease, various early detection methods for urinary tract cancer have been proposed. These methods include screening for hematuria, urinalysis, measurement of biochemical tumor markers in urine and techniques to visualize the urinary tract.

As yet nothing is known of the clinical impact of any type of urinary tract cancer screening in cases of hereditary nonpolyposis colorectal cancer. The present recommendations of the International Collaborative Group on hereditary nonpolyposis colorectal cancer are to screen patients and first degree relatives by sonography and urinalysis starting at age 30 to 35 years at 1 to 2 year intervals, and to limit screening to those families that have manifested urinary tract cancer.[30] This type of screening does not pose much of a physical burden to the patient. However, if other studies confirm our findings that renal cell cancer does not belong to the tumor spectrum of hereditary nonpolyposis colorectal cancer, and risks are highest for renal pelvis and ureteral cancer, then sonography as a screening tool in families is inappropriate, given its limitations in the early detection of renal pelvis and ureteral tumors.[31] Routine urinalysis does not appear to be sensitive in detecting those tumors as well,[32,33] although additional urine screening techniques are being developed to increase detection rates.[34,35] Given the early occurrence of microsatellite instability in the development of hereditary nonpolyposis colorectal cancer tumors in general, looking for this phenomenon in urine specimens[36] might be a useful screening technique. Compared with conventional urine screening, excretory urography (IVP) is more sensitive in detecting transitional cell cancer of the upper urinary tract[37] but it poses a higher burden to the patient and is more expensive. Computerized tomography and magnetic resonance imaging are even more expensive and probably better suited for clinical evaluation of these tumors than for use as

screening tools in asymptomatic subjects.[37] Ureteroscopy is also less suited for screening, given the burden to the patient, including its possible complications.[37]

If one could identify subsets of hereditary nonpolyposis colorectal cancer families with a strong increase in risk of upper urinary tract cancer, then IVP might be justified. In our clinical practice we discuss this type of screening with families with a history of multiple cases of upper urinary tract cancer, especially if it is early onset. Presently, there is insufficient evidence to decide for or against the different types of screening for urinary tract cancer in cases of hereditary nonpolyposis colorectal cancer and, therefore, it should be presented to the families as experimental.

CONCLUSIONS

Our findings indicate that hereditary nonpolyposis colorectal cancer is associated with an increased risk of transitional cell cancer of the upper urinary tract. Although the cumulative risk is relatively low, a subset of families may be exposed to a much higher risk. Future studies should focus on proved carriers of hereditary nonpolyposis colorectal cancer gene mutations to estimate more accurately risks of urinary tract cancer in this disorder. As yet, nothing is known of the clinical impact of screening for urinary tract cancer in cases of hereditary nonpolyposis colorectal cancer. Screening of families with a strong history of upper urinary tract cancer in a research setting by IVP should be considered.

REFERENCES

1. Hodgson, S. V. and Maher, E. R.: A Practical Guide to Human Cancer Genetics. Cambridge: Cambridge University Press, 1993.
2. Kiemeney, L. A. L. M. and Schoenberg, M.: Familial transitional cell carcinoma. J. Urol., **156:** 867, 1996.
3. Lynch, H. T. and Smyrk, T.: Hereditary nonpolyposis colorectal cancer (Lynch syndrome). An updated review. Cancer, **78:** 1149, 1996.
4. Mecklin, J. P. and Järvinen, H. J.: Tumor spectrum in cancer family syndrome (hereditary nonpolyposis colorectal cancer). Cancer, **68:** 1109, 1991.
5. Watson, P. and Lynch, H. T.: Extracolonic cancer in hereditary nonpolyposis colorectal cancer. Cancer, **71:** 677, 1993.
6. Järvinen, H. J., Mecklin, J. P. and Sistonen, P.: Screening reduces colorectal cancer rate in families with hereditary nonpolyposis colorectal cancer. Gastroenterology, **108:** 1405, 1995.
7. Burke, W., Petersen, G., Lynch, P., Botkin, J., Daly, M., Garber, J., Kahn, M. J., McTiernan, A., Offit, K., Thomson, E., Varricchio, C. and the Cancer Genetics Studies Consortium: Recommendations for follow-up care of individuals with an inherited predisposition to cancer: I. Hereditary nonpolyposis colon cancer. J.A.M.A., **277:** 915, 1997.
8. Visser, O., Coebergh, J. W. W., van Leeuwen, F. E. and Schouten, L. J.: Incidence of cancer in The Netherlands, 1990. Second report of The Netherlands Cancer Registry (ISBN 90-72175-07-7). Utrecht: National Association of Comprehensive Cancer Centres, 1994.
9. Breslow, N. E. and Day, N. E.: The Design and Analysis of Cohort Studies. Lyon: IARC, 1987.
10. Zhang, Z.-F. and Steineck, G.: Epidemiology and etiology of bladder cancer. In: Principles and Practice of Genitourinary Oncology. Edited by D. Raghavan, H. I. Scher, S. A. Leibel and P. H. Lange. Philadelphia: Lippincott-Raven, chapt. 19, pp. 215–221, 1997.
11. Aarnio, M., Mecklin, J. P., Aaltonen, L. A., Nyström-Lahti, M. and Järvinen, H. J.: Life-time risk of different cancers in hereditary non-polyposis colorectal cancer (HNPCC) syndrome. Int. J. Cancer, **64:** 430, 1995.
12. Vasen, H. F., Wijnen, J. T., Menko, F. H., Kleibeuker, J. H., Taal, B. G., Griffioen, G., Nagengast, F. M., Meijers-Heijboer, E. H., Bertario, L., Varesco, L., Bisgaard, M. L., Mohr, J., Fodde, R. and Khan, P. M.: Cancer risk in families with hereditary nonpolyposis colorectal cancer diagnosed by mutation analysis. Gastroenterology, **110:** 1020, 1996.
13. Vasen, H. F. A., Mecklin, J. P., Meera Khan, P. and Lynch, H. T.: The International Collaborative Group on HNPCC. Anticancer. Res., **14:** 1661, 1994.
14. Peltomäki, P. and de la Chapelle, A.: Mutations predisposing to hereditary nonpolyposis colorectal cancer. Adv. Cancer. Res., **71:** 93, 1997.
15. Miyaki, M., Konishi, M., Tanaka, K., Kikuchi-Yanoshita, R., Muraoka, M., Yasuno, M., Igari, T., Koike, M., Chiba, M. and Mori, T.: Germline mutation of MSH6 as the cause of hereditary nonpolyposis colorectal cancer. Nat. Genet., **17:** 271, 1997.
16. Rhyu, M. S.: Molecular mechanisms underlying hereditary nonpolyposis colorectal carcinoma. J. Natl. Cancer. Inst., **88:** 240, 1996.
17. Konishi, M., Kikuchi-Yanoshita, R., Tanaka, K., Muraoka, M., Onda, A., Okumura, Y., Kishi, N., Iwama, T., Mori, T., Koike, M., Ushio, K., Chiba, M., Nomizu, S., Konishi, F., Utsunomiya, J. and Miyaki, M.: Molecular nature of colon tumors in hereditary nonpolyposis colon cancer, familial polyposis, and sporadic colon cancer. Gastroenterology, **111:** 307, 1996.
18. Lothe, R. A.: Microsatellite instability in human solid tumors. Mol. Med. Today, **3:** 61, 1997.
19. Gonzalez-Zuleta, M., Ruppert, J. M., Tokino, K., Tsai, Y. C., Spruck, C. H., III, Miyao, N., Nichols, P. W., Hermann, G. G., Horn, T., Steven, K., Summerhayes, I. C., Sidransky, D. and Jones, P. A.: Microsatellite instability in bladder cancer. Cancer. Res., **53:** 5620, 1993.
20. Mao, L., Lee, D. J., Tockman, M. S., Erozan, Y. S., Askin, F. and Sidransky, D.: Microsatellite alterations as clonal markers for the detection of human cancer. Proc. Natl. Acad. Sci., **91:** 9871, 1994.
21. Orlow, I., Lianes, P., Lacombe, L., Dalbagni, G., Reuter, V. E. and Cordon-Cardo, C.: Chromosome 9 allelic losses and microsatellite alterations in human bladder tumors. Cancer Res., **54:** 2848, 1994.
22. Uchida, T., Wada, C., Wang, C., Egawa, S., Ohtani, H. and Koshiba, K.: Genomic instability of microsatellite repeats and mutations of H-, K-, and N-ras, and p53 genes in renal cell carcinoma. Cancer Res., **54:** 3682, 1994.
23. Uchida, T., Wang, C., Wada, C., Iwamura, M., Egawa, S. and Koshiba, K.: Microsatellite instability in transitional cell carcinoma of the urinary tract and its relationship to clinicopathological variables and smoking. Int. J. Cancer, **69:** 142, 1996.
24. Cohen, P. R., Kohn, S. R., Davis, D. A. and Kurzrock, R.: Muir-Torre syndrome. Dermatol. Clin., **13:** 79, 1995.
25. Aaltonen, L. A., Peltomäki, P., Mecklin, J. P., Järvinen, H., Jass, J. R., Green, J. S., Lynch, H. T., Watson, P., Tallqvist, G., Juhola, M., Sistonen, P., Hamilton, S. R., Kinzler, K. W., Vogelstein, B. and de la Chapelle, A.: Replication errors in benign and malignant tumors from hereditary nonpolyposis colorectal cancer patients. Cancer. Res., **54:** 1645, 1994.
26. Honchel, R., Halling, K. C., Schaid, D. J., Pittelkow, M. and Thibodeau, S. N.: Microsatellite instability in Muir-Torre syndrome. Cancer Res., **54:** 1159, 1994.
27. Karsdorp, N., Elderson, A., Wittebol-Post, D., Hené, R. J., Vos, J., Feldberg, M. A., van Gils, A. P., Jansen-Schillhorn van Veen, J. M., Vroom, T. M., Hoppener, J. W. and Lips, C. J. M.: Von Hippel-Lindau disease: new strategies in early detection and treatment. Amer. J. Med., **97:** 158, 1994.
28. Zbar, B., Tory, K., Merino, M., Schmidt, L., Glenn, G., Choyke, P., McClellan, M. W., Lerman, M. and Linehan, W. M.: Hereditary papillary renal cell carcinoma. J. Urol., **151:** 561, 1994.
29. Choyke, P. L., Walther, M. M., Glenn, G. M., Wagner, J. R., Venzon, D. J., Lubensky, I. A., Zbar, B. and Linehan, W. M.: Imaging features of hereditary papillary renal cancers. J. Comput. Asst. Tomogr., **21:** 737, 1997.
30. Weber, T.: Clinical surveillance recommendations adopted for HNPCC. Lancet, **348:** 465, 1996.
31. Hilton, S. and Schwarz, L. H.: Radiologic imaging of genitourinary neoplasms. In: Principles and Practice of Genitourinary Oncology. Edited by D. Raghavan, H. I. Scher, S. A. Leibel and P. H. Lange. Philadelphia: Lippincott-Raven, chapt. 4, pp. 33–42, 1997.
32. Gourlay, W., Chan, V. and Gilks, C. B.: Screening for urothelial malignancies by cytologic analysis and flow cytometry in a community urologic practice: a prospective study. Mod. Path., **8:** 394, 1995.
33. Kern, W. H.: The diagnostic accuracy of sputum and urine cytology. Acta Cytol., **32:** 651, 1988.

34. Parry, W. L. and Hemstreet, G. P., III: Cancer detection by quantitative fluorescence image analysis. J. Urol., **139:** 270, 1988.
35. Ho, K. J. and Kuo, S. H.: Urinary beta-glucuronidase activity as an initial screening test for urinary tract malignancy in high risk patients. Comparison with conventional urine cytologic evaluation. Cancer, **76:** 473, 1995.
36. Mao, L., Schoenberg, M. P., Scicchitano, M., Erozan, Y. S., Merlo, A., Schwab, D. and Sidransky, D.: Molecular detection of primary bladder cancer by microsatellite analysis. Science, **271:** 659, 1996.
37. Brooks, J. D. and Marshall, F. F.: Transitional cell carcinoma of the upper urinary tract. In: Principles and Practice of Genitourinary Oncology. Edited by D. Raghavan, H. I. Scher, S. A. Leibel and P. H. Lange. Philadelphia: Lippincott-Raven, chapt. 32, pp. 337–346, 1997.

Appendix IV
Short Communication: Malignant fibrous histiocytoma in a patient with hereditary non-polyposis colorectal cancer.

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Abstract

Sarcomas, including the malignant fibrous histiocytomas (MFHs), are not part of the tumour spectrum of hereditary non-polyposis colorectal cancer (HNPCC) as epidemiologically established. Therefore, occurrence of MFH in an HNPCC family may very well be coincidental. HNPCC is associated with germline mutations in DNA mismatch repair genes, including the hMSH2 gene. We analyzed an MFH diagnosed in a 45-year-old male hMSH2 mutation carrier for HNPCC-associated molecular characteristics, in order to investigate a possible relationship between the tumour and the germline hMSH2 mutation. DNA analysis revealed microsatellite instability and loss of one hMSH2 copy, immunohistochemistry showed absence of nuclear hMSH2 protein staining. In contrast, in a series of sporadic MFHs, microsatellite instability was not observed in any of the 5 of tumours analyzed and hMSH2 protein staining in the nucleus was demonstrated in all of the 6 tumours tested. Our findings are highly suggestive of a causal relationship in our patient between his hMSH2 germline mutation and the development of his MFH.

Introduction

Hereditary non-polyposis colorectal cancer (HNPCC or Lynch syndrome), is an autosomal dominant cancer predisposition syndrome, associated with a germline mutation in one of five mismatch repair (MMR) genes. Clinical hallmarks are the development of colorectal cancer in 70-90 % of gene mutation carriers, as well as the possible development of a range of extra-colonic tumours including endometrial cancer, ovarian cancer, stomach cancer, small bowel cancer, transitional cell cancer of the upper urinary tract cancer, hepatobiliary cancer and brain tumours[1]. This tumour spectrum has been defined by an epidemiological approach in which the numbers of each of the different types of tumours occurring in HNPCC families were counted and, for risk estimations, were compared with control populations [2-8].

If in an HNPCC patient a tumour occurs which is not included in the recognized HNPCC tumour spectrum, then the question arises whether or not that particular tumour developed as a manifestation of HNPCC. Answering this question may lead to adjusting our knowledge on the HNPCC-associated tumour spectrum and to more insight in the various molecular events which may cause that particular type of tumour.

Occurrence of sarcomas in HNPCC families, including a case of malignant fibrous histiocytoma (MFH)[3], has been reported. However, such findings are rare and may just be coincidental. We report on a male from a large HNPCC family, who was shown to carry a familial germline mutation in the MMR gene hMSH2 and who developed an MFH. To find out whether his germline hMSH2 mutation and his MFH were causally linked, we analyzed his tumour for microsatellite instability, loss of the normal copy of his hMSH2 gene and presence of hMSH2 protein. Because little is known about presence or absence of microsatellite instability and hMSH2 protein expression in sporadic MFHs, we studied a small group of those tumours for comparison.

Report and discussion

The male patient was diagnosed at the age of 45 years with a high-grade MFH of the lower right leg, which was subsequently amputated. From the age of 51 years onward he developed a few colorectal adenomatous polyps with low-grade dysplasia. At the age of 57, an adenocarcinoma of the jejunum was diagnosed. The family of the patient featured multiple patients in successive generations with

colorectal cancer (age at diagnosis 37-48 years), endometrial cancer (40-42 years) or gastric cancer (20-45 years) and met the clinical Amsterdam criteria for HNPCC. The patient as well as his affected relatives, were known with a germline mutation G429X of hMSH2[9].

DNA was extracted from paraffin-embedded specimens of the patient's MFH as well as from five sporadic MFHs. A panel of five microsatellites was tested in these tumours: the two mononucleotide repeats BAT26 and BAT40 and the dinucleotide repeats D2S123, D5S346, S17S250[10]. In the MFH from the HNPCC patient all five markers showed instability, whereas none did in the five sporadic MFHs. Thus, there is a clear indication for a role of a mismatch repair defect in the development of the tumour of the HNPCC patient, whereas no such indication exists for the sporadic MFHs. We tested for loss of heterozygosity (LOH) of the hMSH2 gene in the MFH from the HNPCC patient, using the markers D2S391 and D2S378, located 4cM and 10 CM proximal of the gene, respectively, and D2S2298, which is located 0-3 cM distal of the gene. LOH was observed for all three markers.

To check whether both copies of hMSH2 had been (functionally) eliminated in the tumour, sections of the MFH from the HNPCC patient, and for comparisson also sections of his jejunum adenocarcinoma, were analyzed by immunohistochemistry, using antibodies directed against hMSH2 (Oncogene®, clone FE11, recognizing the carboxy-terminal domain of the protein). As a control, we also used antibodies directed against hMLH1 (Oncogene®, clone G168-728, epitope undetermined). The sections of the MFH of the HNPCC patient were hMLH1 positive, predominantly in the cytoplasm of the tumour cells, and hMSH2 negative (figure 1). In the surrounding connective tissue no positive staining was observed. The sections of the jejunum adenocarcinoma showed hMLH1 and hMSH2 staining in the pre-existent epithelium. The tumour cells were hMSh1 positive and hMSH2 negative (figure 2). Six sporadic MFHs (the five used for MSI testing were not available for immunohistochemistry) were analyzed for comparison. All showed nuclear staining for hMLH1 and hMSH2 (figure 3), one of them also showed cytoplasmatic hMLH1 staining.

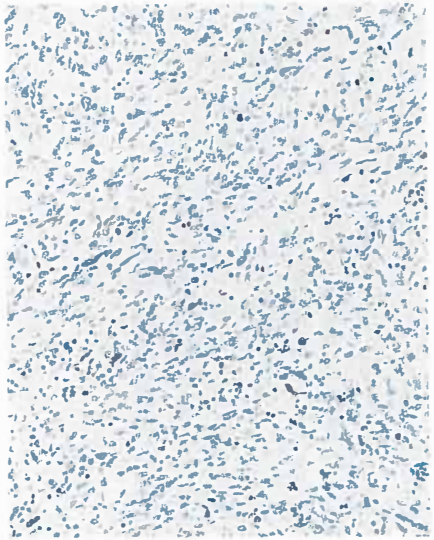

Figure 1

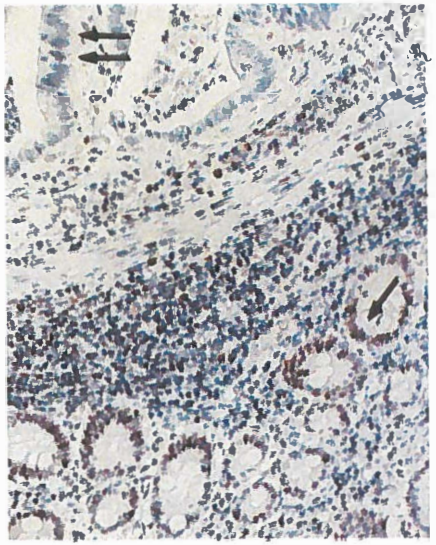

Figure 2

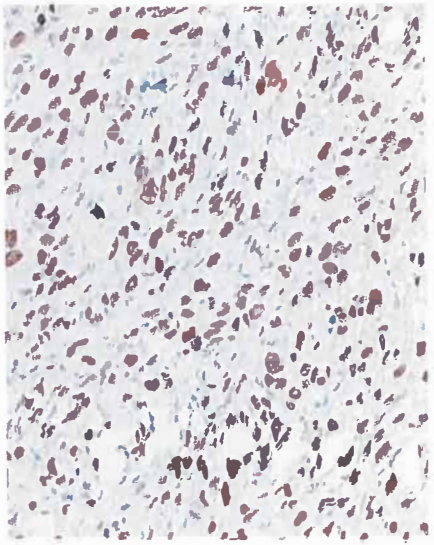

Figure 3

Immunohistochemistry (magnification 250 x). Sections were incubated with the hMSH2 antibody, rabbit-anti-mouse peroxidase (RAMPO) and goat-anti-rabbit peroxidase (GARPO). Diamino-benzidine (DAB) was used as chromogen and haematoxylin as counterstain

Figure 1 *MFH from the HNPCC patient. No staining.*

Figure 2 *Jejunum adenocarcinoma from the HNPCC patient. Bottom: pre-existent epithelium (arrow): positive staining. Top: adenocarcinoma (double-arrow): negative staining.*

Figure 3 *Sporadic MFH. Diffuse positive nuclear staining.*

Our findings strongly suggest that the malignant fibrous histiocytoma which occurred in the HNPCC patient should be regarded as a manifestation of his inherited disorder. Although MSI as observed in his MFH has also been demonstrated in a range of sporadic tumour types[11] and therefore is not proof of HNPCC, MSI seems not to be the rule in sporadic MFHs (ref.[12] and this study).

This might suggest that the mutant hMSH2 allele played a role in the development of the MFH. In addition, we found LOH for markers directly proximal and distal of hMLH2, making it reasonable to assume loss of one of the gene in between. The question remained, however, whether the mutant allele or the wild type allele was lost. This was addressed by the immunohistochemical analysis. In HNPCC associated intestinal cancer, presence of germline hMSH2 gene mutations is strongly associated with negative staining for nuclear hMSH2 protein[13].

Such a negative staining was indeed observed in the jejunum carcinoma from our HNPCC patient. His MFH also stained negative, thus indicating loss of normal hMSH2 protein expression. In contrast, all sporadic MFHs did show hMSH2 staining. However, in the tissue surrounding the MFH in the HNPCC patient we did not observe any positive hMSH2 staining and therefore lack an internal control. This was to be expected, however, given the fibrotic nature of that tissue and knowing that hMSH2 expression is related to proliferative activity (repair of mismatches in replicated DNA). Alternatively, we cannot exclude that fixation conditions prevented staining.

Our findings should alert physicians to the possibility that on rare occasions MFHs may occur in the setting of HNPCC and may be used to search for the MMR gene germline mutations in an HNPCC family where material from relatives with the classical HNPCC associated tumours is unavailable. Survival of patients with HNPCC associated colorectal cancer may differ from that of patients with sporadic cases [14-16], because the molecular etiology of HNPCC-associated tumours may differ from that of non-hereditary cases, as has been demonstrated for colorectal cancer[17-20]. In a similar way, different genes may be involved in MFH development in HNPCC and in the development of sporadic MFH. It cannot be excluded that a difference in survival would, therefore, also apply to patients of both groups of MFH. However, because MFH represents a heterogeneous group of tumours[21], it may very well turn out to be also genetically heterogeneous.

References

1 Lynch HT, Smyrk T, Lynch J. An update of HNPCC (Lynch syndrome). *Cancer Genet Cytogenet* 1997;**93**:84-99.

2 Vasen HFA, Offerhaus GJA, den Hartog-Jager FCA, et al. The tumour spectrum in hereditary non-polyposis colorectal cancer: a study of 24 kindreds in The Netherlands. *Int J Cancer* 1990;**46**:31-34.

3 Mecklin JP, Jarvinen HJ. Tumor spectrum in Cancer Family Syndrome (Hereditary Nonpolyposis Colorectal Cancer). *Cancer* 1991;**68**:1109-1112.

4 Watson P, Lynch HT. Extracolonic cancer in Hereditary Nonpolyposis Colorectal Cancer. *Cancer* 1993;**71**:677-685.

5 Aarnio M, Mecklin JP, Aaltonen LA, Nystrom-Lahti M, Jarvinen HJ. Life-time risk of different cancers in hereditary non- polyposis colorectal cancer (HNPCC) syndrome. *Int J Cancer* 1995;**64**:430-433.

6 Vasen HFA, Wijnen JT, Menko FH, et al. Cancer risk in families with hereditary nonpolyposis colorectal cancer diagnosed by mutation analysis. *Gastroenterology* 1996;**110**:1020-1027.

7 Vasen HFA, Sanders EACM, Taal BG, et al. The risk of brain tumours in hereditary non-polyposis colorectal cancer (HNPCC). *Int J Cancer* 1996;**65**:422-425.

8 Sijmons RH, Kiemeney LALM, Witjes JA, Vasen HFA. Urinary tract cancer and hereditary nonpolyposis colorectal cancer: Risks and screening options. *J Urol* 1998;**160**:466-470.

9 Wijnen J, Vasen H, Meera Khan P, et al. Seven new mutations in hMSH2, an HNPCC gene, identified by denaturing gradient-gel electrophoresis. *Am J Hum Genet* 1995;**56**:1060-1066.

10 Boland CR, Thibodeau SN, Hamilton SR, et al. A National Cancer Institute Workshop on Microsatellite Instability for cancer detection and familial predisposition: development of international criteria for the determination of microsatellite instability in colorectal cancer. *Cancer Res* 1998;**58**:5248-5257.

11 Cunningham JM, Boardman LA, Burgart LJ, Thibodeau SN. Microsatellite instability in colon cancers. In: Wells RD, Warren ST, Sarmiento M, eds. *Genetic Instabilities and Hereditary Neurological Diseases*. San Diego: Academic Press Inc, 1998;791-807.

12 Martin SS, Grear-Hurt W, Hedges LK, Butler MG, Schwartz HS. Microsatellite instability in sarcomas. *Ann Surg Oncol* 1998;**5**:356-360.

13 Fujiwara T, Stolker JM, Watanabe T, et al. Accumulated clonal genetic alterations in familial and sporadic colorectal carcinomas with widespreadinstability in microsatellite sequences. *Am J Pathol* 1998;**153**:1063-1078.

14 Aarnio M, Mustonen H, Mecklin JP, Jarvinen HJ. Prognosis of colorectal cancer varies in different high-risk conditions. *Ann Med* 1998;**30**:75-80.

15 Watson P, Lin KM, Rodriguez-Bigas MA, et al. Colorectal carcinoma survival among hereditary nonpolyposis colorectal carcinoma family members. *Cancer* 1998;**83**:259-266.

16 Bertario L, Russo A, Sala P, et al. Survival of patients with hereditary colorectal cancer: Comparison of HNPCC and colorectal cancer in FAP patients with sporadic colorectal cancer. *Int J Cancer* 1999;**80**:183-187.

17 Konishi M, Kikuchi-Yanoshita R, Tanaka K, et al. Molecular nature of colon tumors in hereditary nonpolyposis colon cancer, familial polyposis, and sporadic colon cancer. *Gastroenterology* 1996;**111**:307-317.

18 Huang J, Papadopoulos N, McKinley AJ, et al. APC mutations in colorectal tumors with mismatch repair deficiency. *Proc Natl Acad Sci USA* 1996;**93**:9049-9054.

19 Tannergard P, Liu T, Weger A, Nordenskjold M, Lindblom A. Tumorigenesis in colorectal tumors from patients with hereditary non- polyposis colorectal cancer. *Hum Genet* 1997;**101**:51-55.

20 Fujiwara T, Stolker JM, Watanabe T, et al. Accumulated clonal genetic alterations in familial and sporadic colorectal carcinomas with widespread instability in microsatellite sequences. *Am J Pathol* 1998;**153**:1063-1078.

21 Meister P. Malignant fibrous histiocytoma: histomorphological pattern or tumor type. *Pathol Res Pract* 1996;**192**:877-881.

Summary and Future Perspectives

Summary

The revolutionary progress in molecular genetics of hereditary cancer is pushing the clinical genetics of cancer into an accelerated development. For a discussion of the issues involved, it is important to clarify the vocabulary. Therefore, in chapter 1 of this thesis a list of definitions is given for frequently used notions in the clinical genetics of cancer. From the review presented in chapter 2 it is clear that many types of cancer may be associated with a human cancer syndrome. To asses the possible presence of one of these syndromes in a cancer patient and his or her family, the family history generally plays a central role. Unfortunately, the contribution of the family history to the genetic diagnosis is limited by its inaccuracy. The study reported in chapter 3 has looked at factors influencing family history accuracy. Its results may help in the interpretation of (as yet) unverified family histories and, if there is no opportunity to verify them all, in deciding which parts of those histories need to be verified.

Recognizing from family histories the clues that point to human cancer syndromes is usually the basis for the cancer-related referral to a clinical genetics centre. However, recognizing these clues may not be easy and some physicians may be better aware of the characteristics of the human cancer syndromes than others. In order to improve the quality of care to families with hereditary cancer it is important to (regionally) distribute and update information on the human cancer syndromes, criteria for referral, options for DNA analysis and preventive measures (*e.g.* screening protocols).

Such actions need, however, to be monitored in order to see whether this transfer of knowledge is successful. In chapter 4 we presented an analysis of the referral from the hospitals and general practitioners in our region.

We observed striking differences within both of these groups and will use the results in a dialogue with all parties involved to identify the cause of these differences and to see whether there is any need to adjust our system of knowledge transfer. This transfer should not be a one-way process. The clinical genetic workload has for an important part shifted in recent years from the traditional topics of congenital anomalies and mental retardation to cancer. Therefore, although non-geneticists may need to learn more about genetic aspects of cancer, clinical geneticists need to learn more about cancer in order to be equal partners in the discussion on hereditary cancer topics.

One way to assist in the recognition of hereditary cancer and in making a clinical genetic differential diagnosis in cancer patients and families, is to present information on this subject in the format of interactive software, as has been done for clinical cytogenetics (the Human Cytogenetics Database by Schinzel), dysmorphology (the London Dysmorphology Database by Winter and Baraitser; POSSUM by Bankier) and some other groups of hereditary disorders. We programmed the Familial Cancer Database (FACD), reported in chapter 5, for this purpose. Only time will tell how much help our programme offers in clinical practice.

Two of the human cancer syndromes, multiple endocrine neoplasia type 2A (MEN2A) and hereditary non-polyposis colorectal cancer (HNPCC) were studied with respect to the disease spectrum and tumour risk. Results are reported in appendices I-IV. In the two MEN2A studies, patients with familial cutaneous lichen amyloidosis (CLA) and familial or isolated Hirschsprung disease (HSCR), respectively, were examined for mutations in the RET gene (which is associated with MEN2). The reason for this was that both disorders have also been reported to occur in MEN2A patients. We therefore questioned whether CLA and HSCR patients without a MEN2A family history could carry MEN2A-associated RET mutations and thus could have an increased cancer risk. Indeed, such mutations were observed in HSCR patients. Although tumour risks for these HSCR patients are as yet difficult to estimate, our study underlines the fact that some patients with disorders known to be associated with human cancer syndromes, may carry mutant genes associated with an increased cancer risk in the absence of a family history fitting those syndromes. Physicians diagnosing and treating patients with such disorders should be aware of this phenomenon in the interest of tumour prevention.

Disease spectrum and tumour risk in HNPCC were explored in the studies reported in appendices III and IV. The first study used an epidemiological approach to look at the association of cancer of the urinary tract with HNPCC in all HNPCC families

included in the National HNPCC Registry maintained by the Netherlands Foundation for the Detection of Hereditary Tumours and presents risk figures for these tumours. Studies such as this one have the advantage of generating risk figures, but cannot learn us much of the nature of tumours which occur only rarely in a human cancer syndrome.

The second study demonstrates a totally different approach to study the disease spectrum. Using molecular techniques, a single case of malignant fibrous histiocytoma (very rarely occurring in HNPCC) diagnosed in an HNPCC patient was tested for molecular characteristics of HNPCC. The results suggested that this tumour had indeed developed as part of HNPCC in this patient. Both types of study, each with its own particular advantages and disadvantages, are needed to further delineate the disease spectrum of the human cancer syndromes.

Future Perspectives

Many questions relevant to the clinical genetics of cancer remain to be answered. Some of these may be more readily answered than others. For example, it is unlikely that we will be able in the near future to precisely predict the clinical implications of a mutant gene in individual cases. Inter- and intra-familial differences in phenotypical expression of one and the same mutant gene are the rule rather than the exception in human cancer syndromes and can be explained by the multi-step nature of cancer development and the fact that a wide range of external as well as internal risk and protective factors may interact in complex and therefore largely unpredictable ways.

The future will, however, undoubtedly see an expansion of diagnostic capabilities. In particular, we can expect DNA analysis to contribute increasingly to the genetic diagnosis in patients and families suspected of having a human cancer syndrome. Although increased mutation detection rates will generate a further increase in demand for genetic services with all the practical budgetary problems tied in with this increase, the possibility to detect all mutations associated with human cancer syndromes will also alleviate the burden of family history verification and the 'wrestling' with differential diagnosis.

Questions with regard to tumour prevention in individuals and families will probably take many more years to solve, which implies that more people will be diagnosed with an inherited strong cancer predisposition at a time when the value of preventive options will still be very unclear for many of the human cancer syndromes as reviewed in chapter 2. Only large collaborative studies can address questions with

respect to outcome of periodic screening, tumour incidence after prophylactic surgery and the short- and long-range effects and side-effects of chemoprevention. This implies that the (preventive) treatment and follow-up of patients should be protocolized in order to be able to compare and pool data. This is a strong argument in favour of centralising these medical activities. However, in practice, many of these activities are not performed in central settings.

Although genetic analysis is often centralised per region because of the very specialised expertise involved (and because of legislation, as in our country), depending on the choice of inclusion criteria, the load of patients for further medical management may be too large to cope with in a central clinic. Also, patients may prefer to remain in the care of specialists working outside such central clinics. If decentralised medical management would follow the mentioned protocols and its local registration could be carried out in a way that would allow for the pooling of data on regional and preferably national levels so that a study population could be defined large enough for meaningful statistical analysis, then that could be an adequate alternative. Nevertheless, certain medical procedures (*e.g.* prophylactic thyroidectomy in small children, presymptomatic DNA testing including its psychological support) should remain centralised for the present in order to guarantee expertise.

Looking at the future, it can hardly be expected that diagnostic and even presymptomatic DNA testing and genetic counselling will forever remain the exclusive domain of clinical genetics centres. Simply because of the sheer number of patients involved and the opening of international markets for commercial DNA testing, decentralisation of testing and counselling seems likely. The role of clinical genetics centres may shift towards one which mainly focuses on hereditary disorders which are particularly complex with regard to DNA testing, risk analysis and counselling, as well as on disorders for which, because of their rare nature, commercial testing is unavailable.

With respect to the other disorders, an increasing role as a consultancy service, rather than as a service which performs all of the actual testing and counselling, may be expected. However, these changes should not be encouraged until comprehensive mutation analysis will be possible and the clinical and psychosocial implications of the results from such analyses are further clarified.

Samenvatting

Klinische genetica is het medische specialisme dat zich bezighoudt met het onderzoek naar erfelijke en aangeboren aandoeningen, ook wel erfelijkheidsonderzoek genoemd en de voorlichting van patiënt en familie over deze aandoeningen, ook wel erfelijkheidsvoorlichting genoemd. Dit proefschrift beschrijft een aantal onderzoeken op het gebied van de klinische genetica van kanker. Voordat deze onderzoeken hier worden samengevat volgt eerst een korte uitleg over wat kanker en erfelijkheid wel en niet met elkaar te maken hebben.

Cellen, genen, erfelijke ziekten en kanker

Ons lichaam is opgebouwd uit miljarden bouwstenen, die cellen worden genoemd. Ze zijn er in alle soorten en maten: huidcellen, spiercellen, zenuwcellen, bloedcellen en nog vele anderen. Elke cel bevat een "keuken" waar van alles en nog wat "gekookt" wordt aan de hand van "recepten" die klaarliggen in die cel. Deze recepten zijn stukjes van ons erfelijke materiaal, ons DNA. De kookresultaten zijn de *eiwitten*. Cellen hebben eiwitten nodig om hun taken kunnen uitvoeren.

Elk recept wordt een *gen* genoemd. Genen komen in cellen in tweevoud voor, waarbij het ene exemplaar van de moeder en het andere van de vader afkomstig is. Genen kunnen beschadigd raken. Ouders kunnen via hun ei- of zaadcellen (waarin de genen in enkelvoud voorkomen) beschadigde genen doorgeven aan hun kinderen. Als een gen beschadigd is, dan wordt er aan de hand van een afwijkend recept "gekookt", waardoor een ziekte kan ontstaan. Een ziekte, die het gevolg is van een geërfde beschadiging, noemen we *erfelijk* en het beschadigde gen, de *aanleg* voor die erfelijke ziekte.

Het is ook mogelijk dat in de loop van het leven in gewone lichaamscellen beschadigingen ontstaan in genen die onbeschadigd van de ouders waren geërfd. Sommigen genen bevatten informatie die belangrijk is voor controle van het delen van cellen, ze functioneren als het ware als "gaspedalen" en "remmen" voor de celdeling. Een bepaalde opeenstapeling van beschadigingen van deze genen leidt ertoe dat een cel de controle op zijn celdeling verliest.

Ongecontroleerde celgroei wordt in het lichaam uiteindelijk zichtbaar als een gezwel. Als een gezwel doorgroeit in omgevende weefsels en zich uitzaait naar andere delen van het lichaam, dan noemen we dat gezwel kwaadaardig, ofwel *kanker*. Sommige genbeschadigingen werken het ontstaan van kanker sterk in de hand, andere geven slechts een "zwak zetje" in die richting.

De opeenstapeling van beschadigingen van genen die tot het ontstaan van kanker leidt, ontstaat in normaal erfelijk materiaal, meestal in de loop van vele jaren, door de inwerking op deze genen van bepaalde chemische stoffen en straling. Ook ontstaat deze schade voor een gedeelte spontaan door foutjes tijdens celdelingsprocessen. Omdat al deze beschadigingen niet geërfd zijn, noemen we kanker in dit geval dan ook geen erfelijke ziekte.

In ongeveer 1 tot 10 % van alle gevallen van kanker is er echter wel sprake van een geërfde genbeschadiging die belangrijk bij heeft gedragen aan het ontstaan van die kanker. In deze gevallen spreken we over *erfelijke kanker*. Het ziektebeeld dat hoort bij een bepaalde aanleg wordt in die gevallen *erfelijk kanker syndroom* (hereditary cancer syndrome) of *humaan kanker syndroom* (human cancer syndrome) genoemd. Er bestaan tientallen van die syndromen. De meest voorkomende zijn erfelijke borst-eierstokkanker en het erfelijk non-polyposis colorectaal carcinoom, een erfelijke vorm van darm- en baarmoederkanker.

Samenvatting

In **hoofdstuk 1** worden definities gegeven van de begrippen die belangrijk zijn voor de klinische genetica van kanker en wordt in een notendop geschetst wat het erfelijkheidsonderzoek naar kanker en de erfelijkheidsvoorlichting inhouden. Daarnaast worden de algemene kenmerken van de erfelijke kankersyndromen beschreven.

In **hoofdstuk 2** wordt van elk van de meest voorkomende soorten kanker (uitgezonderd het non-Hodgkin lymfoom en leukemie) beschreven of het voorkomen van die soort bij iemand de kans voor bloedverwanten op kanker verhoogt en meer in het bijzonder, of die soort voor kan komen als onderdeel van een erfelijke kankersyndroom. In veel gevallen van kanker zal het voor een arts niet onmiddellijk duidelijk zijn dat het bij zijn patiënt wel of niet om een erfelijke vorm gaat. Het maken van een onderscheid tussen een erfelijke en niet erfelijke vorm kan van groot belang zijn voor de behandeling en preventieve controles bij

patiënt en familieleden. Er wordt daarom ingegaan op kenmerken van erfelijke kanker die als herkenningssignaal kunnen dienen in de medische praktijk. Het voorkomen van kanker bij meerdere familieleden is daar één van. Omdat kanker veel voorkomt zullen alleen al door toeval meerdere gevallen van kanker in een familie kunnen voorkomen. Het voorkomen van kanker in een familie bewijst dus niet dat die kanker erfelijk is. Daarom is het letten op andere kenmerken van erfelijke kanker ook van belang, zoals relatief jonge leeftijd bij ontstaan, alsook vorming van verschillende gezwellen bij één en dezelfde persoon (uitzaaiingen worden daarbij niet meegeteld). Sommige combinaties van kanker in een familie wijzen sterk op een erfelijke vorm. De combinaties borst/eierstokkanker en darm/baarmoeder (niet baarmoederhals) kanker zijn de meest voorkomende voorbeelden hiervan. Tenslotte wordt in dit hoofdstuk ingegaan op de huidige kennis over de preventieve maatregelen die men kan nemen om erfelijke kanker te voorkomen of zo vroeg mogelijk op te sporen. Geconcludeerd wordt, dat van veel van deze maatregelen de voor- en nadelen nog niet duidelijk vaststaan.

Als mensen vragen om erfelijkheidsonderzoek naar kanker wordt hen gevraagd gegevens te verstrekken over de kanker bij zichzelf en hun familieleden. Het is belangrijk dat deze gegevens kloppen, omdat de klinisch geneticus er sterk op vaart bij het nemen van beslissingen over de mogelijkheden voor DNA onderzoek, het stellen van de genetische diagnose en het verwijzen voor preventieve controles en eventueel operaties. Als regel worden de familiegegevens daarom voor alle zekerheid geverifieerd en wel aan de hand van informatie die wordt opgevraagd bij ziekenhuizen en huisartsen. Dit is echter een tijdrovend en in sommige gevallen zelfs onmogelijk karwei, bijvoorbeeld als de medische gegevens inmiddels vernietigd zijn of omdat toestemming voor het opvragen van gegevens ontbreekt. In **hoofdstuk 3** wordt een onderzoek beschreven naar de betrouwbaarheid van familiegegevens. Hieruit kwam naar voren dat verreweg de meeste door de familie gemelde gevallen van borstkanker accuraat waren. Voor meldingen van bijvoorbeeld baarmoederkanker was dat veel minder het geval. Er wordt geconcludeerd dat het minder verifiëren van met name meldingen over borstkanker, niet noemenswaard ten koste gaat van de juistheid van de genetische diagnose, mits dit volgens een bepaalde schema verloopt. Het minder verifiëren van familiegegevens levert een aanzienlijke tijdsbesparing op.

De belangstelling bij zowel het publiek als artsen voor de erfelijke aspecten van kanker is de afgelopen jaren sterk toegenomen en daarmee ook de vraag naar genetisch onderzoek. In **hoofdstuk 4** werd dit geïnventariseerd. In Groningen groeide de vraag om erfelijkheidsonderzoek voor kanker van 11 vragen per jaar in 1987 naar meer dan 300 per jaar in 1997. Op dit moment gaan 30 tot 40 % van al onze verzoeken om erfelijkheidsonderzoek over kanker, terwijl dat in 1987 slechts 3 % was. Hoewel de vraag naar erfelijkheidsonderzoek voor kanker enorm is toegenomen, is het de vraag of iedereen die in aanmerking komt voor dat onderzoek en dat onderzoek ook zou willen, daadwerkelijk verwezen wordt. Het is best mogelijk dat sommige verwijzers nog niet goed op de hoogte zijn van de criteria voor verwijzing. Mogelijk moeten de verwijscriteria en de voor- en nadelen van erfelijkheidsonderzoek naar kanker nog beter onder de aandacht worden gebracht van sommige (potentiële) verwijzers.

Om voorlichting over dit onderwerp beter te kunnen richten, werden de verwijspatronen van de regionale huisartsen en de specialisten van de 17 regionale niet-academische ziekenhuizen in kaart gebracht. Omdat regio's en ziekenhuizen onderling verschillen in het aantal patiënten dat met bepaalde soorten kanker gezien wordt, moesten de verwijzingen naar ons centrum voor die verschillen gecorrigeerd worden.

Met name voor borst en eierstokkanker blijken er tussen ziekenhuizen duidelijke verschillen in verwijzing te bestaan en dat geldt ook voor huisartsen als we die in subregio's indelen. Wat precies de oorzaken zijn voor de gevonden verschillen en of nu inderdaad een manier is gevonden om bijscholing te richten, moet nog blijken uit vervolgonderzoek.

In **hoofdstuk 5** wordt het computerprogramma Familial Cancer Database beschreven, dat werd ontwikkeld als geheugensteun voor artsen bij het herkennen van erfelijke vormen van kanker. De gebruiker van het programma kan de ziektekenmerken invoeren, in het bijzonder de soorten kanker, die bij een patiënt en familieleden werden gevonden. Het programma geeft dan vervolgens een overzicht van alle erfelijke aandoeningen waarbij die ziektekenmerken kunnen voorkomen. Op dit moment bevat Familial Cancer Database gegevens over meer dan 300 verschillende aandoeningen. Een groot voordeel van het gebruik van een computerprogramma is, dat het snel kan worden aangepast aan nieuwe kennis over de verschillende erfelijke aandoeningen.

In de **aanhangsels I tot en met IV** komen twee erfelijke kankersyndromen aan bod: multipele endocriene neoplasie type 2a (MEN2A) en hereditair non-polyposis colorectaal carcinoom (HNPCC). Patiënten met MEN2A hebben een bepaalde soort schildklierkanker en vaak ook bijnier- en bijschildkliergezwellen. Soms gaat MEN2A gepaard met een bepaalde huidaandoening (cutane lichen amyloidosis, CLA) of een bepaalde aangeboren afwijking van de zenuwen van de darm (de ziekte van Hirschsprung). Patiënten met HNPCC hebben meestal darm- en/of baarmoederkanker, maar ook andere soorten kanker kunnen bij hen voorkomen.

Appendix I beschrijft DNA onderzoek in families met zowel MEN2A als CLA en in families waarin alleen CLA voorkomt. De conclusie is dat CLA zonder MEN2A hoogstwaarschijnlijk een aandoening is die niets met de aanleg voor MEN2A te maken heeft. De bevindingen bij mensen met de ziekte van Hirschsprung zonder MEN2A, zoals die beschreven worden in **appendix II**, zijn echter anders. In familiaire of geïsoleerd voorkomende gevallen van deze darmziekte kan wel degelijk een MEN2A aanleg worden gevonden. Dit betekent dat deze patiënten op latere leeftijd alsnog verschijnselen van MEN2A kunnen gaan vertonen. Behandelaars dienen hierop alert te zijn.

In **appendix III** is voor Nederlandse families met HNPCC uitgerekend of ze naast een hoge kans op darm- en baarmoederkanker, ook een verhoogde kans op kanker van de urinewegen hebben. Dit blijkt inderdaad het geval te zijn voor nierbekken- en urineleiderkanker, alhoewel deze kansen duidelijk lager zijn dan de kans op darm- en baarmoederkanker.

In **appendix IV** werd onderzocht of het gezwel dat een HNPCC patiënt in zijn been kreeg, een zogenaamd maligne fibreus histiocytoom, iets te maken had met zijn HNPCC aanleg of niet. Met behulp van DNA onderzoek en het gebruik van speciale antistoffen op weefselcoupes (immunohistochemie) kon een verband met deze aanleg inderdaad aannemelijk worden gemaakt.

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Curriculum Vitae

Rolf (Roelof Han) Sijmons graduated in medicine in 1986 (Groningen). He served as a conscript-physician at the National Army 477 Regional Medical Unit between 1986 and 1987. From 1987 to 1989 he worked at the Radiation Oncology Department (chair at that time: Prof.dr.J.Vermeij) of the University Hospital in Groningen. In 1989 he joined the Department of Medical Genetics (chair: Prof.dr.C.H.C.M.Buys) of the University of Groningen. He trained in clinical genetics under Prof.dr.L.P.ten Kate and obtained his specialist degree in 1993. His current fields of interest are hereditary cancer and clinical cytogenetics.

Publications

Sijmons, R.H. Aetiology and epidemiology - report of rapporteur. In: Weber, W. ed. *Familial Cancer Control*. Berlin: Springer Verlag, 1992, p. 35-36.

De Boer,M., Bolscher, B.G., Sijmons, R.H., Scheffer, H., Weening, R.S., Roos,D. Prenatal diagnosis in a family with X-linked chronic granulomatous disease with the use of the polymerase chain reaction. *Prenat Diagn* 1992;12(9): 773-7.

Sijmons, R.H., Vasen, H.F.A. Groeiende aandacht voor erfelijke kanker. *Kanker* 1992;16:170-173.

Sijmons, R.H., Leegte, B., van Lingen, R.A., de Pater, J.M., van der Veen, A.Y., del Canho, H., Bos, C., ten Kate, L.P., Breed, A.S. Tetrasomy 5p mosaicism in a boy with delayed growth, hypotonia, minor anomalies, and an additional isochromosome 5p [46,XY/47,XY, + i(5p)]. *Am J Med Genet* 47(4): 559-62, 1993.

Sijmons, R.H., Kristoffersson,U., Tuerlings,J.H., Ljung,R., Dijkhuis-Stoffelsma,R.; Breed, A.S. Piebaldism in a mentally retarded girl with rare deletion of the long arm of chromosome 4. *Pediatr Dermatol* 10(3): 235-9, 1993.

Sijmons, R.H. Steeds meer bekend over erfelijke vormen van kanker. *Kanker* 1993;17:232-233.

Oosterwijk, J.C., Sijmons, R.H., Menko, F.H., Chorus, A.M.J., and Rookus, M.A. De kans op mammacarcinoom en samenhangende carcinomen bij een positieve familieanamnese. *Ned Tijdschr Geneeskd* 1995;139:423-428.

Sijmons, R.H., Boonstra, H., and Vasen, H.F.A. Erfelijke aspecten van gynaecologische tumoren. *Ned Tijdschrift Obst Gynaecol* 1995;108:411-412.

de Walle H.E.K., Cornel M.C., Sijmons R.H., Tuerlings J.H.A.M., Kate L.P.t. Overlevingskansen van kinderen met het Down syndroom in Noord-Nederland. *Tijdschr Kindergeeskd* 1995;**2**:40-44.

Sijmons R.H. Erfelijke borstkanker: de praktijk. *Huisartsen College* 1995;**4**:6-7.

Sijmons R.H., Sikkema-Raddatz B., Kloosterman M.D., Briet J.W., Jong B.d., Leschot N.J. 46,XY,dup(10q) in direct CVS preparation and mosaic 48,XXXY, dup(10q) in CVS long-term culture and fetal tissue. *Prenat Diagn* 1995;**15**:285-290.

Sijmons R.H., van Essen A.J., Visser J.D., Iprenburg M., Nelck G.F., Vos-Bender M.L. Congenital knee dislocation in a 49,XXXXY boy. *J Med Genet* 1995;**32**:309-311.

Sikkema-Raddatz B., Sijmons R.H., Tan-Sindhunata M.B., Veen A.Y.v.d., Brunsting R., de Vries B., et al. Prenatal diagnosis in two cases of de novo complex balanced chromosomal rearrangements. Three-year follow-up in one case. *Prenat Diagn* 1995;**15**:467-473.

Hofstra, R.M.W., Sijmons, R.H., Stelwagen, T., Stulp, R.P., Kousseff, B.G., Lips, C.J.M., Steijlen, P.M., van Voorst Vader, P.C., and Buys, C.H.C.M. RET mutation screening in familial cutaneous lichen amyloidosis and in skin amyloidosis associated with multiple endocrine neoplasia. *J Invest Dermatol* 1996;107(2):215-218.

Schaap, C., Sijmons, R.H. Toenemende inzichten bij erfelijke vormen van kanker. *Kanker* 1996;20:16-19.

Sijmons, R.H., Sikkens, E.H., and Grosfeld, F.J.M. Presymptomatic DNA testing in families with multiple endocrine neoplasia type 2A: Practical counseling issues. In: Muller, H., Scott, R.J., Weber, W, eds. *Hereditary Cancer*, Basel: Karger, 1996, p. 191-195.

Boonstra, H., Sijmons, R.H. Het endometriumcarcinoom bij het hereditaire nonpolyposis colorectaal carcinoom: een update van het 'Cancer Family Syndrome' oftewel het Lynch II syndroom. *Ned Tijdschrift Obst Gynaecol* 1997;110:34-36.

Nelen, M.R., van Staveren, W.C.G., Peeters, E.A.J., Ben Hassel, M., Gorlin, R.J., Hamm, H., Lindboe, C.F., Fryns, J.P., Sijmons, R.H., Woods, D.G., Mariman, E.C.M., Padberg, G.W., and Kremer, H. Germline mutations in the PTEN/MMAC1 gene in patients with Cowden disease. *Hum Mol Genet* 1997;6(8):1383-1387.

Peelen, T., van Vliet, M., Petrij-Bosch, A., Mieremet, R., Szabo, C., van den Ouweland, A.M.W., Hogervorst, F., Brohet, R., Ligtenberg, M.J.L., Teugels, E., Van der Luijt, R., van der Hout, A.H., Gille, J.J.P., Pals, G., Jedema, I., Olmer, R., van Leeuwen, I., Newman, B., Plandsoen, M., van der Est, M., Brink, G., Hageman, S., Arts, P.J.W., Bakker, M.M., Willems, H.W., van der Looij, E., Neyns, B., Bonduelle, M., Jansen, R., Oosterwijk, J.C., Sijmons, R., Smeets, H.J.M., van Asperen, C.J., Meijers-Heijboer, H., Klijn, J.G.M., de Greve, J., King, M.C., Menko, F.H., Brunner, H.G., Halley, D., van Ommen, G.J.B., Vasen, H.F.A., Cornelisse, C.J., van 't Veer, L.J., de Knijff, P., Bakker, E., and Devilee, P. A high proportion of novel mutations in BRCA1 with strong founder effects among Dutch and Belgian hereditary breast and ovarian cancer families. *Am J Hum Genet* 1997;60(5):1041-1049.

Kroes, H.Y., Sijmons, R.H., Van den Berg, A., van der Hout, A.H. Early-onset renal cell cancer and bilateral epididymal cysts as presenting symptoms of von Hippel-Lindau disease. *Br J Urol* 1998;81:915.

Menko, F.H., Wijnen, J.T., Vasen, H.F.A., Sijmons, R.H., and Meera Khan, P. Familial and hereditary non-polyposis colorectal cancer: issues relevant for surgical practice. *Cancer Res* 1998;146:20-31.

Sikkema-Raddatz, B., Sijmons, R.H. De novo balanced complex chromosomal rearrangements in prenatal diagnosis. In: Rao, K.A. and Nicolaides, K eds. *Chorionic villus sampling*, New Delhi: Jaypee Brothers, 1998, p. 99-103.

Wijnen, J.T., Vasen, H.F.A., Meera Khan, P., Zwinderman, A.H., Van der Klift, H., Mulder, A., Tops, C., Moller, P., Fodde, R., Menko, F., Taal, B., Nagengast, F., Brunner, H., Kleibeuker, J., Sijmons, R., Griffioen, G., Brocker-Vriends, A., Bakker, E., Van Leeuwen-Cornelisse, I., Meijers-Heijboer, A., Lindhout, D., Breuning, M., Post, J., Schaap, C., Apold, J., Heimdal, K., Bertario, L., Bisgaard, M.L., and Goetz, P. Clinical findings with implications for genetic testing in families with clustering of colorectal cancer. *N Engl J Med* 1998;339(8):511-518.

Sijmons, R.H., Hofstra, R.M.W., Wijburg, F.A., Links, T.P., Zwierstra, R.P., Vermey, A., Aronson, D.C., Tan-Sindhunata, G., Brouwers-Smalbraak, G.J., Maas, S.M., and Buys, C.H.C.M. Oncological implications of RET gene mutations in Hirschsprung's disease. *Gut* 1998;43:542-547.

Sijmons, R.H., Kiemeney, L.A.L.M., Witjes, J.A., and Vasen, H.F.A. Urinary tract cancer and hereditary nonpolyposis colorectal cancer: risks and screening options. *J Urol* 1998;160(2):466-470.